(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
24 March 2005 (24.03.2005)

PCT

(10) International Publication Number
WO 2005/026370 A2

(51) International Patent Classification[7]: C12N 15/86

(21) International Application Number:
PCT/US2004/028751

(22) International Filing Date:
3 September 2004 (03.09.2004)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

60/500,572	5 September 2003 (05.09.2003)	US
60/504,007	18 September 2003 (18.09.2003)	US

(71) Applicants *(for all designated States except US)*: **AVENTIS PASTEUR LIMTED** [CA/CA]; 1755 Steeles Avenue West, Toronto, Ontario M2R 3T4 (CA). **THERION BIOLOGICS, INC.** [US/US]; 76 Rodgers Street, Cambridge, MA 02142-1119 (US).

(72) Inventors; and

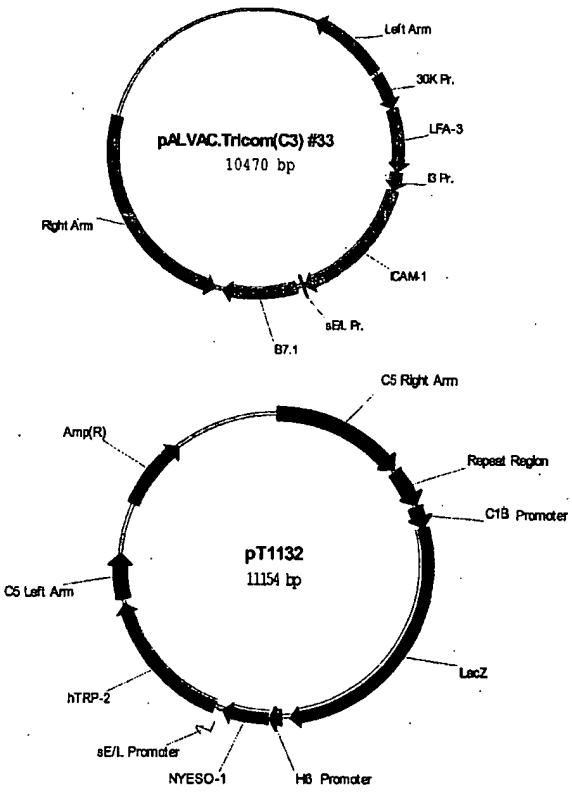
(75) Inventors/Applicants *(for US only)*: **BERINSTEIN, Neil** [CA/CA]; Aventis Pasteur Limited, 1755 Steeles Avenue West, Toronto, Ontario M2R 3T4 (CA). **TARTAGLIA, Jim** [CA/CA]; Aventis Pasteur Limited, 1755 Steeles Avenue West, Toronto, Ontario M2R 3T4 (CA). **PARRINGTON, Mark** [CA/CA]; Aventis Pasteur Limited, 1755 Steeles Avenue West, Toronto, Ontario M2R 3T4 (CA). **PANICALI, Dennis** [US/US]; 76 Rogers Street, Cambridge, MA 02142-1119 (US). **GRITZ, Linda** [US/US]; 76 Rogers Street, Cambridge, MA 02142-1119 (US).

(74) Agent: **HALLORAN, Patrick, J.**; Aventis Pasteur, Discovery Drive, Swiftwater, PA 18370 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI,

[Continued on next page]

(54) Title: MULTI-ANTIGEN VECTORS FOR MELANOMA



(57) Abstract: The present invention relates to peptides, polypeptides, and nucleic acids and the use of the peptide, polypeptide or nucleic acid in preventing and / or treating cancer. In particular, the invention relates to peptides and nucleic acid sequences encoding such peptides for use in diagnosing, treating, or preventing melanoma.

WO 2005/026370 A2

GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) **Designated States** *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— *without international search report and to be republished upon receipt of that report*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

Multi-Antigen Vectors for Melanoma

FIELD OF THE INVENTION

The present invention relates to multi-antigen vectors for use in preventing and / or treating cancer. In particular, the invention relates to multi-antigen vectors for use in treating and/or preventing melanoma.

BACKGROUND OF THE INVENTION

There has been tremendous increase in last few years in the development of cancer vaccines with tumour-associated antigens (TAAs) due to the great advances in identification of molecules based on the expression profiling on primary tumours and normal cells with the help of several techniques such as high density microarray, SEREX, immunohistochemistry (IHC), RT-PCR, in-situ hybridization (ISH) and laser capture microscopy (Rosenberg, Immunity, 1999; Sgroi et al, 1999, Schena et al, 1995, Offringa et al, 2000). The TAAs are antigens expressed or over-expressed by tumour cells and could be specific to one or several tumours for example CEA antigen is expressed in colorectal, breast and lung cancers. Sgroi et al (1999) identified several genes differentially expressed in invasive and metastatic carcinoma cells with combined use of laser capture microdissection and cDNA microarrays. Several delivery systems like DNA or viruses could be used for therapeutic vaccination against human cancers (Bonnet et al, 2000) and can elicit immune responses and also break immune tolerance against TAAs. Tumour cells can be rendered more immunogenic by inserting transgenes encoding T cell co-stimulatory molecules such as B7.1 or cytokines such as IFN-γ, IL2, or GM-CSF, among others. Co-expression of a TAA and a cytokine or a co-stimulatory molecule can develop effective therapeutic vaccine (Hodge et al, 95, Bronte et al, 1995, Chamberlain et al, 1996).

There is a need in the art for reagents and methodologies useful in stimulating an immune response to prevent or treat cancers. The present invention provides such reagents and methodologies that overcome many of the difficulties encountered by others in attempting to treat cancer.

SUMMARY OF THE INVENTION

The present invention provides multi-antigen vectors for administration to a patient to prevent and / or treat cancer. In particular, the multi-antigen vector encodes one or more tumor antigens ("TA"). The multi-antigen vector may also encode an immune stimulator such as a co-stimulatory molecule and/or be administered with an adjuvant.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1. Schematic of plasmids pALVAC.Tricom(#33) and pT1132.

Figure 2. DNA sequence of plasmid pALVAC.Tricom(#33).

Figure 3. DNA sequence of plasmid pT1132.

Figure 4. Schematic of plasmid pT3217.

Figure 5. DNA sequence of plasmid pT3217.

Figure 6. Amino acid sequences of exemplary NY-ESO-1, TRP-2, gp100, gp100M, MART-1, MAGE-1, MAGE-3, B7.1, LFA-3, and ICAM-1 proteins.

DETAILED DESCRIPTION

The present invention provides reagents and methodologies useful for treating and / or preventing cancer. All references cited within this application are incorporated by reference.

In one embodiment, the present invention relates to the induction or enhancement of an immune response against one or more tumor antigens ("TA") to prevent and / or treat cancer. In certain embodiments, one or more TAs may be combined. In preferred embodiments, the immune response results from expression of a TA in a host cell following administration of a nucleic acid vector encoding the tumor antigen or the tumor antigen itself in the form of a peptide or polypeptide, for example.

As used herein, an "antigen" is a molecule (such as a polypeptide) or a portion thereof that produces an immune response in a host to whom the antigen has been administered. The immune response may include the production of antibodies that bind to at least one epitope of the antigen and / or the generation of a cellular immune response against cells expressing an epitope of the antigen. The response may be an enhancement of a current immune response by, for example, causing increased antibody production, production of antibodies with increased affinity for the antigen, or an increase in the cellular immune response (i.e., increased number or activity

of immunoreactive T cells). An antigen that produces an immune response may alternatively be referred to as being immunogenic or as an immunogen. In describing the present invention, a TA may be referred to as an "immunogenic target". The present invention provide expression vectors for expressing in a host one or more immunogenic targets.

The term TA includes both tumor-associated antigens (TAAs) and tumor-specific antigens (TSAs), where a cancerous cell is the source of the antigen. A TAA is an antigen that is expressed on the surface of a tumor cell in higher amounts than is observed on normal cells or an antigen that is expressed on normal cells during fetal development. A TSA is an antigen that is unique to tumor cells and is not expressed on normal cells. TA further includes TAAs or TSAs, antigenic fragments thereof, and modified versions that retain their antigenicity.

TAs are typically classified into five categories according to their expression pattern, function, or genetic origin: cancer-testis (CT) antigens (i.e., MAGE, NY-ESO-1); melanocyte differentiation antigens (i.e., Melan A/MART-1, tyrosinase, gp100); mutational antigens (i.e., MUM-1, p53, CDK-4); overexpressed 'self' antigens (i.e., HER-2/neu, p53); and, viral antigens (i.e., HPV, EBV). For the purposes of practicing the present invention, a suitable TA is any TA that induces or enhances an anti-tumor immune response in a host to whom the TA has been administered. Suitable TAs include, for example, species of gp100 (Cox et al., *Science*, 264:716-719 (1994); U.S. Pat. No. 6,500,919 B1 and WO 01/30847 with Val at residue 162, also referred to as "gp100M"; U.S. Pat. No. 6,537,560 B1 with Phe at residue 162), MART-1/Melan A (Kawakami et al., *J. Exp. Med.*, 180:347-352 (1994); U.S. Pat. No. 5,874,560), gp75 (TRP-1) (Wang et al., *J. Exp. Med.*, 186:1131-1140 (1996)), TRP-2 (Wang et al. 1996 J. Exp. Med. 184:2207; U.S. Pat. Nos. 5,831,016 and 6,083,783), tyrosinase (Wolfel et al., *Eur. J. Immunol.*, 24:759-764 (1994); WO 200175117; WO 200175016; WO 200175007), NY-ESO-1 (WO 98/14464; WO 99/18206; GenBank Accession No. P78358; U.S. Pat. No. 5,804,381), melanoma proteoglycan (Hellstrom et al., *J. Immunol.*, 130:1467-1472 (1983)), MAGE family antigens (i.e., MAGE-1, 2,3,4,6,12, 51; Van der Bruggen et al., *Science*, 254:1643-1647 (1991); U.S. Pat. Nos. 6,235,525; CN 1319611), BAGE family antigens (Boel et al., *Immunity*, 2:167-175 (1995)), GAGE family antigens (i.e., GAGE-1,2; Van den Eynde et al., *J. Exp. Med.*, 182:689-698 (1995); U.S. Pat. No. 6,013,765), RAGE family antigens (i.e., RAGE-1; Gaugler et at., *Immunogenetics*, 44:323-330 (1996); U.S. Pat. No. 5,939,526), N-acetylglucosaminyltransferase-V (Guilloux et at., *J. Exp. Med.*, 183:1173-1183 (1996)), p15 (Robbins et al., *J. Immunol.*

154:5944-5950 (1995)), ß-catenin (Robbins et al., *J. Exp. Med.*, 183:1185-1192 (1996)), MUM-1 (Coulie et al., *Proc. Natl. Acad. Sci.* USA, 92:7976-7980 (1995)), cyclin dependent kinase-4 (CDK4) (Wolfel et al., *Science*, 269:1281-1284 (1995)), p21-*ras* (Fossum et at., *Int. J. Cancer*, 56:40-45 (1994)), BCR-*abl* (Bocchia et al., *Blood*, 85:2680-2684 (1995)), p53 (Theobald et al., *Proc. Natl. Acad. Sci.* USA, 92:11993-11997 (1995)), p185 HER2/neu (erb-B1; Fisk et al., *J. Exp. Med.*, 181:2109-2117 (1995)), epidermal growth factor receptor (EGFR) (Harris et al., Breast Cancer Res. Treat, 29:1-2 (1994)), carcinoembryonic antigens (CEA) (Kwong et al., *J. Natl. Cancer Inst.*, 85:982-990 (1995) U.S. Pat. Nos. 5,756,103; 5,274,087; 5,571,710; 6,071,716; 5,698,530; 6,045,802; EP 263933; EP 346710; and, EP 784483); carcinoma-associated mutated mucins (i.e., MUC-1 gene products; Jerome et al., *J. Immunol.*, 151:1654-1662 (1993)); EBNA gene products of EBV (i.e., EBNA-1; Rickinson et al., *Cancer Surveys*, 13:53-80 (1992)); E7, E6 proteins of human papillomavirus (Ressing et al., *J. Immunol*, 154:5934-5943 (1995)); prostate specific antigen (PSA; Xue et al., *The Prostate*, 30:73-78 (1997)); prostate specific membrane antigen (PSMA; Israeli, et al., *Cancer Res.*, 54:1807-1811 (1994)); idiotypic epitopes or antigens, for example, immunoglobulin idiotypes or T cell receptor idiotypes (Chen et al., *J. Immunol.*, 153:4775-4787 (1994)); KSA (U.S. Patent No. 5,348,887), kinesin 2 (Dietz, et al. Biochem Biophys Res Commun 2000 Sep 7;275(3):731-8), HIP-55, TGFβ-1 anti-apoptotic factor (Toomey, et al. Br J Biomed Sci 2001;58(3):177-83), tumor protein D52 (Bryne J.A., et al., Genomics, 35:523-532 (1996)), H1FT, NY-BR-1 (WO 01/47959), NY-BR-62, NY-BR-75, NY-BR-85, NY-BR-87, NY-BR-96 (Scanlan, M. Serologic and Bioinformatic Approaches to the Identification of Human Tumor Antigens, in *Cancer Vaccines 2000*, Cancer Research Institute, New York, NY), including "wild-type" (i.e., normally encoded by the genome, naturally-occurring), modified, and mutated versions as well as other fragments and derivatives thereof. Any of these TAs may be utilized alone or in combination with one another in a co-immunization protocol.

Preferred TAs are useful for inducing an immune response against melanoma cells. The term "melanoma" includes but is not limited to melanomas, metastatic melanomas, melanomas derived from either melanocytes or melanocyte related nevus cells, melanocarcinomas, melanoepitheliomas, melanosarcomas, melanoma *in situ*, superficial spreading melanoma, nodular melanoma, lentigo maligna melanoma, acral lentiginous melanoma, invasive melanoma and familial atypical mole and melanoma (FAM-M) syndrome, for example. In general,

melanomas result from chromosomal abnormalities, degenerative growth and development disorders, mitogenic agents, ultraviolet radiation (UV), viral infections, inappropriate tissue expression of a gene, alterations in expression of a gene or carcinogenic agents, for example.

In certain cases, it may be beneficial to co-immunize patients with both TA and other antigens, such as angiogenesis-associated antigens ("AA"). An AA is an immunogenic molecule (i.e., peptide, polypeptide) associated with cells involved in the induction and / or continued development of blood vessels. For example, an AA may be expressed on an endothelial cell ("EC"), which is a primary structural component of blood vessels. Where the cancer is cancer, it is preferred that that the AA be found within or near blood vessels that supply a tumor. Immunization of a patient against an AA preferably results in an anti-AA immune response whereby angiogenic processes that occur near or within tumors are prevented and / or inhibited. Exemplary AAs include, for example, vascular endothelial growth factor (i.e., VEGF; Bernardini, et al. *J. Urol.*, 2001, 166(4): 1275-9; Starnes, et al. *J. Thorac. Cardiovasc. Surg.*, 2001, 122(3): 518-23; Dias, et al. *Blood*, 2002, 99: 2179-2184), the VEGF receptor (i.e., VEGF-R, flk-1/KDR; Starnes, et al. *J. Thorac. Cardiovasc. Surg.*, 2001, 122(3): 518-23), EPH receptors (i.e., EPHA2; Gerety, et al. 1999, *Cell*, 4: 403-414), epidermal growth factor receptor (i.e., EGFR; Ciardeillo, et al. Clin. Cancer Res., 2001, 7(10): 2958-70), basic fibroblast growth factor (i.e., bFGF; Davidson, et al. Clin. Exp. Metastasis 2000,18(6): 501-7; Poon, et al. Am J. Surg., 2001, 182(3):298-304), platelet-derived cell growth factor (i.e., PDGF-B), platelet-derived endothelial cell growth factor (PD-ECGF; Hong, et al. J. Mol. Med., 2001, 8(2):141-8), transforming growth factors (i.e., TGF-α; Hong, et al. J. Mol. Med., 2001, 8(2):141-8), endoglin (Balza, et al. *Int. J. Cancer*, 2001, 94: 579-585), Id proteins (Benezra, R. Trends Cardiovasc. Med., 2001, 11(6):237-41), proteases such as uPA, uPAR, and matrix metalloproteinases (MMP-2, MMP-9; Djonov, et al. J. Pathol., 2001, 195(2):147-55), nitric oxide synthase (Am. J. Ophthalmol., 2001, 132(4):551-6), aminopeptidase (Rouslhati, E. Nature Cancer, 2: 84-90, 2002), thrombospondins (i.e., TSP-1, TSP-2; Alvarez, et al. Gynecol. Oncol., 2001, 82(2):273-8; Seki, et al. Int. J. Oncol., 2001, 19(2):305-10), k-*ras* (Zhang, et al. Cancer Res., 2001, 61(16):6050-4), *Wnt* (Zhang, et al. Cancer Res., 2001, 61(16):6050-4), cyclin-dependent kinases (CDKs; Drug Resist. Updat. 2000, 3(2):83-88), microtubules (Timar, et al. 2001. *Path. Oncol. Res.*, 7(2): 85-94), heat shock proteins (i.e., HSP90 (Timar, *supra*)), heparin-binding factors (i.e., heparinase; Gohji, et al. Int. J. Cancer, 2001, 95(5):295-301), synthases (i.e., ATP synthase,

thymidilate synthase), collagen receptors, integrins (i.e., αυβ3, αυβ5, α1β1, α2β1, α5β1), the surface proteolglycan NG2, AAC2-1, or AAC2-2, among others, including "wild-type" (i.e., normally encoded by the genome, naturally-occurring), modified, mutated versions as well as other fragments and derivatives thereof. Any of these targets may be suitable in practicing the present invention, either alone or in combination with one another or with other agents.

The nucleic acid molecule may comprise or consist of a nucleotide sequence encoding one or more immunogenic targets, or fragments or derivatives thereof, such as that contained in a DNA insert in an ATCC Deposit. The term "nucleic acid sequence" or "nucleic acid molecule" refers to a DNA or RNA sequence. The term encompasses molecules formed from any of the known base analogs of DNA and RNA such as, but not limited to 4-acetylcytosine, 8-hydroxy-N6-methyladenosine, aziridinyl-cytosine, pseudoisocytosine, 5-(carboxyhydroxylmethyl) uracil, 5-fluorouracil, 5-bromouracil, 5-carboxymethylaminomethyl-2-thiouracil, 5-carboxymethylaminomethyluracil, dihydrouracil, inosine, N6-iso-pentenyladenine, 1-methyladenine, 1-methylpseudouracil, 1-methylguanine, 1-methylinosine, 2,2-dimethyl-guanine, 2-methyladenine, 2-methylguanine, 3-methylcytosine, 5-methylcytosine, N6-methyladenine, 7-methylguanine, 5-methylaminomethyluracil, 5-methoxyamino-methyl-2-thiouracil, beta-D-mannosylqueosine, 5' -methoxycarbonyl-methyluracil, 5-methoxyuracil, 2-methylthio-N6-isopentenyladenine, uracil-5-oxyacetic acid methylester, uracil-5-oxyacetic acid, oxybutoxosine, pseudouracil, queosine, 2-thiocytosine, 5-methyl-2-thiouracil, 2-thiouracil, 4-thiouracil, 5-methyluracil, N-uracil-5-oxyacetic acid methylester, uracil-5-oxyacetic acid, pseudouracil, queosine, 2-thiocytosine, and 2,6-diaminopurine, among others.

An isolated nucleic acid molecule is one that: (1) is separated from at least about 50 percent of proteins, lipids, carbohydrates, or other materials with which it is naturally found when total nucleic acid is isolated from the source cells; (2) is not be linked to all or a portion of a polynucleotide to which the nucleic acid molecule is linked in nature; (3) is operably linked to a polynucleotide which it is not linked to in nature; and / or, (4) does not occur in nature as part of a larger polynucleotide sequence. Preferably, the isolated nucleic acid molecule of the present invention is substantially free from any other contaminating nucleic acid molecule(s) or other contaminants that are found in its natural environment that would interfere with its use in polypeptide production or its therapeutic, diagnostic, prophylactic or research use. As used herein, the term "naturally occurring" or "native" or "naturally found" when used in connection

with biological materials such as nucleic acid molecules, polypeptides, host cells, and the like, refers to materials which are found in nature and are not manipulated by man. Similarly, "non-naturally occurring" or "non-native" as used herein refers to a material that is not found in nature or that has been structurally modified or synthesized by man.

The identity of two or more nucleic acid or amino acid sequences is determined by comparing the sequences. As known in the art, "identity" means the degree of sequence relatedness between nucleic acid or amino acid sequences as determined by the match between the units making up the molecules (i.e., nucleotides or amino acid residues). Identity measures the percent of identical matches between the smaller of two or more sequences with gap alignments (if any) addressed by a particular mathematical model or computer program (i.e., an algorithm). Identity between nucleic acid sequences may also be determined by the ability of the nucleic acid sequences to hybridize to one another. In defining the process of hybridization, the term "highly stringent conditions" and "moderately stringent conditions" refer to conditions that permit hybridization of nucleic acid strands whose sequences are complementary, and to exclude hybridization of significantly mismatched nucleic acids. Examples of "highly stringent conditions" for hybridization and washing are 0.015 M sodium chloride, 0.0015 M sodium citrate at 65-68°C or 0.015 M sodium chloride, 0.0015 M sodium citrate, and 50% formamide at 42°C. (see, for example, Sambrook, Fritsch & Maniatis, *Molecular Cloning: A Laboratory Manual* (2nd ed., Cold Spring Harbor Laboratory, 1989); Anderson *et al.*, *Nucleic Acid Hybridisation: A Practical Approach* Ch. 4 (IRL Press Limited)). The term "moderately stringent conditions" refers to conditions under which a DNA duplex with a greater degree of base pair mismatching than could occur under "highly stringent conditions" is able to form. Exemplary moderately stringent conditions are 0.015 M sodium chloride, 0.0015 M sodium citrate at 50-65°C or 0.015 M sodium chloride, 0.0015 M sodium citrate, and 20% formamide at 37-50°C. By way of example, moderately stringent conditions of 50°C in 0.015 M sodium ion will allow about a 21% mismatch. During hybridization, other agents may be included in the hybridization and washing buffers for the purpose of reducing non-specific and/or background hybridization. Examples are 0.1% bovine serum albumin, 0.1% polyvinyl-pyrrolidone, 0.1% sodium pyrophosphate, 0.1% sodium dodecylsulfate, $NaDodSO_4$, (SDS), ficoll, Denhardt's solution, sonicated salmon sperm DNA (or another non-complementary DNA), and dextran sulfate, although other suitable agents can also be used. The concentration and types of these

additives can be changed without substantially affecting the stringency of the hybridization conditions.. Hybridization experiments are usually carried out at pH 6.8-7.4; however, at typical ionic strength conditions, the rate of hybridization is nearly independent of pH.

In preferred embodiments of the present invention, vectors are used to transfer a nucleic acid sequence encoding an immunogenic target to a cell. A vector is any molecule used to transfer a nucleic acid sequence to a host cell. In certain cases, an expression vector is utilized. An expression vector is a nucleic acid molecule that is suitable for transformation of a host cell and contains nucleic acid sequences that direct and / or control the expression of the transferred nucleic acid sequences. Expression includes, but is not limited to, processes such as transcription, translation, and splicing, if introns are present. Expression vectors typically comprise one or more flanking sequences operably linked to a heterologous nucleic acid sequence encoding a polypeptide. Flanking sequences may be homologous (i.e., from the same species and / or strain as the host cell), heterologous (i.e., from a species other than the host cell species or strain), hybrid (i.e., a combination of flanking sequences from more than one source), or synthetic, for example.

A flanking sequence is preferably capable of effecting the replication, transcription and / or translation of the coding sequence and is operably linked to a coding sequence. As used herein, the term operably linked refers to a linkage of polynucleotide elements in a functional relationship. For instance, a promoter or enhancer is operably linked to a coding sequence if it affects the transcription of the coding sequence. However, a flanking sequence need not necessarily be contiguous with the coding sequence, so long as it functions correctly. Thus, for example, intervening untranslated yet transcribed sequences can be present between a promoter sequence and the coding sequence and the promoter sequence may still be considered operably linked to the coding sequence. Similarly, an enhancer sequence may be located upstream or downstream from the coding sequence and affect transcription of the sequence.

In certain embodiments, it is preferred that the flanking sequence is a transcriptional regulatory region that drives high-level gene expression in the target cell. The transcriptional regulatory region may comprise, for example, a promoter, enhancer, silencer, repressor element, or combinations thereof. The transcriptional regulatory region may be either constitutive, tissue-specific, cell-type specific (i.e., the region is drives higher levels of transcription in a one type of tissue or cell as compared to another), or regulatable (i.e., responsive to interaction with a

compound such as tetracycline). The source of a transcriptional regulatory region may be any prokaryotic or eukaryotic organism, any vertebrate or invertebrate organism, or any plant, provided that the flanking sequence functions in a cell by causing transcription of a nucleic acid within that cell. A wide variety of transcriptional regulatory regions may be utilized in practicing the present invention.

Suitable transcriptional regulatory regions include the CMV promoter (i.e., the CMV-immediate early promoter); promoters from eukaryotic genes (i.e., the estrogen-inducible chicken ovalbumin gene, the interferon genes, the gluco-corticoid-inducible tyrosine aminotransferase gene, and the thymidine kinase gene); and the major early and late adenovirus gene promoters; the SV40 early promoter region (Bernoist and Chambon, 1981, *Nature* 290:304-10); the promoter contained in the 3' long terminal repeat (LTR) of Rous sarcoma virus (RSV) (Yamamoto, *et al.*, 1980, *Cell* 22:787-97); the herpes simplex virus thymidine kinase (HSV-TK) promoter (Wagner *et al.*, 1981, *Proc. Natl. Acad. Sci. U.S.A.* 78:1444-45); the regulatory sequences of the metallothionine gene (Brinster *et al.*, 1982, *Nature* 296:39-42); prokaryotic expression vectors such as the beta-lactamase promoter (Villa-Kamaroff *et al.*, 1978, *Proc. Natl. Acad. Sci. U.S.A.*, 75:3727-31); or the tac promoter (DeBoer *et al.*, 1983, *Proc. Natl. Acad. Sci. U.S.A.*, 80:21-25). Tissue- and / or cell-type specific transcriptional control regions include, for example, the elastase I gene control region which is active in pancreatic acinar cells (Swift *et al.*, 1984, *Cell* 38:639-46; Ornitz *et al.*, 1986, *Cold Spring Harbor Symp. Quant. Biol.* 50:399-409 (1986); MacDonald, 1987, *Hepatology* 7:425-515); the insulin gene control region which is active in pancreatic beta cells (Hanahan, 1985, *Nature* 315:115-22); the immunoglobulin gene control region which is active in lymphoid cells (Grosschedl *et al.*, 1984, *Cell* 38:647-58; Adames *et al.*, 1985, *Nature* 318:533-38; Alexander *et al.*, 1987, *Mol. Cell. Biol.*, 7:1436-44); the mouse mammary tumor virus control region in testicular, breast, lymphoid and mast cells (Leder *et al.*, 1986, *Cell* 45:485-95); the albumin gene control region in liver (Pinkert *et al.*, 1987, *Genes and Devel.* 1:268-76); the alpha-feto-protein gene control region in liver (Krumlauf *et al.*, 1985, *Mol. Cell. Biol.*, 5:1639-48; Hammer *et al.*, 1987, *Science* 235:53-58); the alpha 1-antitrypsin gene control region in liver (Kelsey *et al.*, 1987, *Genes and Devel.* 1:161-71); the beta-globin gene control region in myeloid cells (Mogram *et al.*, 1985, *Nature* 315:338-40; Kollias *et al.*, 1986, *Cell* 46:89-94); the myelin basic protein gene control region in oligodendrocyte cells in the brain (Readhead *et al.*, 1987, *Cell* 48:703-12); the myosin light chain-2 gene control region in

skeletal muscle (Sani, 1985, *Nature* 314:283-86); the gonadotropic releasing hormone gene control region in the hypothalamus (Mason *et al.*, 1986, *Science* 234:1372-78), and the tyrosinase promoter in melanoma cells (Hart, I. Semin Oncol 1996 Feb;23(1):154-8; Siders, et al. Cancer Gene Ther 1998 Sep-Oct;5(5):281-91), among others. Inducible promoters that are activated in the presence of a certain compound or condition such as light, heat, radiation, tetracycline, or heat shock proteins, for example, may also be utilized (see, for example, WO 00/10612). Other suitable promoters are known in the art.

As described above, enhancers may also be suitable flanking sequences. Enhancers are cis-acting elements of DNA, usually about 10-300 bp in length, that act on the promoter to increase transcription. Enhancers are typically orientation- and position-independent, having been identified both 5' and 3' to controlled coding sequences. Several enhancer sequences available from mammalian genes are known (i.e., globin, elastase, albumin, alpha-feto-protein and insulin). Similarly, the SV40 enhancer, the cytomegalovirus early promoter enhancer, the polyoma enhancer, and adenovirus enhancers are useful with eukaryotic promoter sequences. While an enhancer may be spliced into the vector at a position 5' or 3' to nucleic acid coding sequence, it is typically located at a site 5' from the promoter. Other suitable enhancers are known in the art, and would be applicable to the present invention.

While preparing reagents of the present invention, cells may need to be transfected or transformed. Transfection refers to the uptake of foreign or exogenous DNA by a cell, and a cell has been transfected when the exogenous DNA has been introduced inside the cell membrane. A number of transfection techniques are well known in the art (i.e., Graham *et al.*, 1973, *Virology* 52:456; Sambrook *et al.*, *Molecular Cloning, A Laboratory Manual* (Cold Spring Harbor Laboratories, 1989); Davis *et al.*, *Basic Methods in Molecular Biology* (Elsevier, 1986); and Chu *et al.*, 1981, *Gene* 13:197). Such techniques can be used to introduce one or more exogenous DNA moieties into suitable host cells.

In certain embodiments, it is preferred that transfection of a cell results in transformation of that cell. A cell is transformed when there is a change in a characteristic of the cell, being transformed when it has been modified to contain a new nucleic acid. Following transfection, the transfected nucleic acid may recombine with that of the cell by physically integrating into a chromosome of the cell, may be maintained transiently as an episomal element without being

replicated, or may replicate independently as a plasmid. A cell is stably transformed when the nucleic acid is replicated with the division of the cell.

The expression vectors of the present invention also provide for expression of fragments of immunogenic targets. Fragments may include sequences truncated at the amino terminus (with or without a leader sequence) and / or the carboxy terminus. Fragments may also include variants (i.e., allelic, splice), orthologs, homologues, and other variants having one or more amino acid additions or substitutions or internal deletions as compared to the parental sequence. In preferred embodiments, truncations and/or deletions comprise about 1-5 amino acids, 5-10 amino acids, 10-20 amino acids, 20-30 amino acids, 30-40 amino acids, 40-50 amino acids, or more. Such polypeptide fragments may optionally comprise an amino terminal methionine residue. It will be appreciated that such fragments can be used, for example, to generate antibodies or cellular immune responses to immunogenic targets.

A variant is a sequence having one or more sequence substitutions, deletions, and/or additions as compared to the subject sequence. Variants may be naturally occurring or artificially constructed. Such variants may be prepared from the corresponding nucleic acid molecules. In preferred embodiments, the variants have from 1 to 3, or from 1 to 5, or from 1 to 10, or from 1 to 15, or from 1 to 20, or from 1 to 25, or from 1 to 30, or from 1 to 40, or from 1 to 50, or more than 50 amino acid substitutions, insertions, additions and/or deletions.

An allelic variant is one of several possible naturally-occurring alternate forms of a sequence occupying a given locus on a chromosome of an organism or a population of organisms. A splice variant is a polypeptide generated from one of several RNA transcript resulting from splicing of a primary transcript. An ortholog is a similar nucleic acid or polypeptide sequence from another species. For example, the mouse and human versions of an immunogenic target may be considered orthologs of each other. A derivative of a sequence is one that is derived from a parental sequence those sequences having substitutions, additions, deletions, or chemically modified variants. Variants may also include fusion proteins, which refers to the fusion of one or more first sequences (such as a peptide) at the amino or carboxy terminus of at least one other sequence (such as a heterologous peptide).

"Similarity" is a concept related to identity, except that similarity refers to a measure of relatedness which includes both identical matches and conservative substitution matches. If two polypeptide sequences have, for example, 10/20 identical amino acids, and the remainder are all

non-conservative substitutions, then the percent identity and similarity would both be 50%. If in the same example, there are five more positions where there are conservative substitutions, then the percent identity remains 50%, but the percent similarity would be 75% (15/20). Therefore, in cases where there are conservative substitutions, the percent similarity between two polypeptides will be higher than the percent identity between those two polypeptides.

Substitutions may be conservative, or non-conservative, or any combination thereof. Conservative amino acid modifications to the sequence of a polypeptide (and the corresponding modifications to the encoding nucleotides) may produce polypeptides having functional and chemical characteristics similar to those of a parental polypeptide. For example, a "conservative amino acid substitution" may involve a substitution of a native amino acid residue with a non-native residue such that there is little or no effect on the size, polarity, charge, hydrophobicity, or hydrophilicity of the amino acid residue at that position and, in particlar, does not result in decreased immunogenicity. Suitable conservative amino acid substitutions are shown in **Table I.**

Table I

Original Residues	Exemplary Substitutions	Preferred Substitutions
Ala	Val, Leu, Ile	Val
Arg	Lys, Gln, Asn	Lys
Asn	Gln	Gln
Asp	Glu	Glu
Cys	Ser, Ala	Ser
Gln	Asn	Asn
Glu	Asp	Asp
Gly	Pro, Ala	Ala
His	Asn, Gln, Lys, Arg	Arg
Ile	Leu, Val, Met, Ala, Phe, Norleucine	Leu
Leu	Norleucine, Ile, Val, Met, Ala, Phe	Ile
Lys	Arg, 1,4 Diamino-butyric Acid, Gln, Asn	Arg
Met	Leu, Phe, Ile	Leu
Phe	Leu, Val, Ile, Ala, Tyr	Leu
Pro	Ala	Gly
Ser	Thr, Ala, Cys	Thr
Thr	Ser	Ser
Trp	Tyr, Phe	Tyr
Tyr	Trp, Phe, Thr, Ser	Phe
Val	Ile, Met, Leu, Phe, Ala, Norleucine	Leu

A skilled artisan will be able to determine suitable variants of an immunogenic target using well-known techniques. For identifying suitable areas of the molecule that may be changed without destroying biological activity (i.e., MHC binding, immunogenicity), one skilled in the art may target areas not believed to be important for that activity. For example, when immunogenic targets with similar activities from the same species or from other species are known, one skilled in the art may compare the amino acid sequence of a polypeptide to such similar polypeptides. By performing such analyses, one can identify residues and portions of the molecules that are conserved. It will be appreciated that changes in areas of the molecule that are not conserved relative to such similar immunogenic targets would be less likely to adversely affect the biological activity and/or structure of a polypeptide. Similarly, the residues required for binding to MHC are known, and may be modified to improve binding. However, modifications resulting in decreased binding to MHC will not be appropriate in most situations. One skilled in the art would also know that, even in relatively conserved regions, one may substitute chemically similar amino acids for the naturally occurring residues while retaining activity. Therefore, even areas that may be important for biological activity or for structure may be subject to conservative amino acid substitutions without destroying the biological activity or without adversely affecting the structure of the immunogenic target.

Other preferred polypeptide variants include glycosylation variants wherein the number and/or type of glycosylation sites have been altered compared to the subject amino acid sequence. In one embodiment, polypeptide variants comprise a greater or a lesser number of N-linked glycosylation sites than the subject amino acid sequence. An N-linked glycosylation site is characterized by the sequence Asn-X-Ser or Asn-X-Thr, wherein the amino acid residue designated as X may be any amino acid residue except proline. The substitution of amino acid residues to create this sequence provides a potential new site for the addition of an N-linked carbohydrate chain. Alternatively, substitutions that eliminate this sequence will remove an existing N-linked carbohydrate chain. Also provided is a rearrangement of N-linked carbohydrate chains wherein one or more N-linked glycosylation sites (typically those that are naturally occurring) are eliminated and one or more new N-linked sites are created. To affect O-linked glycosylation of a polypeptide, one would modify serine and / or threonine residues.

Additional preferred variants include cysteine variants, wherein one or more cysteine residues are deleted or substituted with another amino acid (*e.g.*, serine) as compared to the subject amino acid sequence set. Cysteine variants are useful when peptides or polypeptides must be refolded into a biologically active conformation such as after the isolation of insoluble inclusion bodies. Cysteine variants generally have fewer cysteine residues than the native protein, and typically have an even number to minimize interactions resulting from unpaired cysteines.

In other embodiments, the peptides or polypeptides may be attached to one or more fusion segments that assist in purification of the polypeptides. Fusions can be made either at the amino terminus or at the carboxy terminus of the subject polypeptide variant thereof. Fusions may be direct with no linker or adapter molecule or may be through a linker or adapter molecule. A linker or adapter molecule may be one or more amino acid residues, typically from about 20 to about 50 amino acid residues. A linker or adapter molecule may also be designed with a cleavage site for a DNA restriction endonuclease or for a protease to allow for the separation of the fused moieties. It will be appreciated that once constructed, the fusion polypeptides can be derivatized according to the methods described herein. Suitable fusion segments include, among others, metal binding domains (e.g., a poly-histidine segment), immunoglobulin binding domains (i.e., Protein A, Protein G, T cell, B cell, Fc receptor, or complement protein antibody-binding domains), sugar binding domains (e.g., a maltose binding domain), and/or a "tag" domain (i.e., at least a portion of α-galactosidase, a strep tag peptide, a T7 tag peptide, a FLAG peptide, or other domains that can be purified using compounds that bind to the domain, such as monoclonal antibodies). This tag is typically fused to the peptide or polypeptide and upon expression may serve as a means for affinity purification of the sequence of interest polypeptide from the host cell. Affinity purification can be accomplished, for example, by column chromatography using antibodies against the tag as an affinity matrix. Optionally, the tag can subsequently be removed from the purified sequence of interest polypeptide by various means such as using certain peptidases for cleavage. As described below, fusions may also be made between a TA and a co-stimulatory components such as the chemokines CXC10 (IP-10), CCL7 (MCP-3), or CCL5 (RANTES), for example.

A fusion motif may enhance transport of an immunogenic target to an MHC processing compartment, such as the endoplasmic reticulum. These sequences, referred to as tranduction or

transcytosis sequences, include sequences derived from HIV tat (see Kim et al. 1997 J. Immunol. 159:1666), *Drosophila* antennapedia (see Schutze-Redelmeier et al. 1996 J. Immunol. 157:650), or human period-1 protein (hPER1; in particular, SRRHHCRSKAKRSRHH).

In addition, the polypeptide or variant thereof may be fused to a homologous peptide or polypeptide to form a homodimer or to a heterologous peptide or polypeptide to form a heterodimer. Heterologous peptides and polypeptides include, but are not limited to an epitope to allow for the detection and/or isolation of a fusion polypeptide; a transmembrane receptor protein or a portion thereof, such as an extracellular domain or a transmembrane and intracellular domain; a ligand or a portion thereof which binds to a transmembrane receptor protein; an enzyme or portion thereof which is catalytically active; a polypeptide or peptide which promotes oligomerization, such as a leucine zipper domain; a polypeptide or peptide which increases stability, such as an immunoglobulin constant region; a peptide or polypeptide which has a therapeutic activity different from the peptide or polypeptide; and/or variants thereof.

In certain embodiments, it may be advantageous to combine a nucleic acid sequence encoding an immunogenic target with one or more co-stimulatory component(s) such as cell surface proteins, cytokines or chemokines in a composition of the present invention. The co-stimulatory component may be included in the composition as a polypeptide or as a nucleic acid encoding the polypeptide, for example. Suitable co-stimulatory molecules include, for instance, polypeptides that bind members of the CD28 family (i.e., CD28, ICOS; Hutloff, et al. *Nature* 1999, 397: 263–265; Peach, et al. *J Exp Med* 1994, 180: 2049–2058) such as the CD28 binding polypeptides B7.1 (CD80; Schwartz, 1992; Chen et al, 1992; Ellis, et al. *J. Immunol.*, 156(8): 2700-9), B7.2 (CD86; Ellis, et al. *J. Immunol.*, 156(8): 2700-9), and mutants / variants thereof (WO 00/66162); polypeptides which bind members of the integrin family (i.e., LFA-1 (CD11a / CD18); Sedwick, et al. *J Immunol* 1999, 162: 1367–1375; Wülfing, et al. *Science* 1998, 282: 2266–2269; Lub, et al. *Immunol Today* 1995, 16: 479–483) including members of the ICAM family (i.e., ICAM-1, -2 or -3); polypeptides which bind CD2 family members (i.e., CD2, signalling lymphocyte activation molecule (CDw150 or "SLAM"; Aversa, et al. *J Immunol* 1997, 158: 4036–4044)) such as CD58 (LFA-3; CD2 ligand; Davis, et al. *Immunol Today* 1996, 17: 177–187) or SLAM ligands (Sayos, et al. *Nature* 1998, 395: 462–469); polypeptides which bind heat stable antigen (HSA or CD24; Zhou, et al. *Eur J Immunol* 1997, 27: 2524–2528); polypeptides which bind to members of the TNF receptor (TNFR) family (i.e.,

4-1BB (CD137; Vinay, et al. *Semin Immunol* 1998, 10: 481–489), OX40 (CD134; Weinberg, et al. *Semin Immunol* 1998, 10: 471–480; Higgins, et al. *J Immunol* 1999, 162: 486–493), and CD27 (Lens, et al. *Semin Immunol* 1998, 10: 491–499)) such as 4-1BBL (4-1BB ligand; Vinay, et al. *Semin Immunol* 1998, 10: 481–48; DeBenedette, et al. *J Immunol* 1997, 158: 551–559), TNFR associated factor-1 (TRAF-1; 4-1BB ligand; Saoulli, et al. *J Exp Med* 1998, 187: 1849–1862, Arch, et al. *Mol Cell Biol* 1998, 18: 558–565), TRAF-2 (4-1BB and OX40 ligand; Saoulli, et al. *J Exp Med* 1998, 187: 1849–1862; Oshima, et al. *Int Immunol* 1998, 10: 517–526, Kawamata, et al. *J Biol Chem* 1998, 273: 5808–5814), TRAF-3 (4-1BB and OX40 ligand; Arch, et al. *Mol Cell Biol* 1998, 18: 558–565; Jang, et al. *Biochem Biophys Res Commun* 1998, 242: 613–620; Kawamata S, et al. *J Biol Chem* 1998, 273: 5808–5814), OX40L (OX40 ligand; Gramaglia, et al. *J Immunol* 1998, 161: 6510–6517), TRAF-5 (OX40 ligand; Arch, et al. *Mol Cell Biol* 1998, 18: 558–565; Kawamata, et al. *J Biol Chem* 1998, 273: 5808–5814), and CD70 (CD27 ligand; Couderc, et al. *Cancer Gene Ther.*, 5(3): 163-75). CD154 (CD40 ligand or "CD40L"; Gurunathan, et al. *J. Immunol.*, 1998, 161: 4563-4571; Sine, et al. *Hum. Gene Ther.*, 2001, 12: 1091-1102) may also be suitable.

One or more cytokines may also be suitable co-stimulatory components or "adjuvants", either as polypeptides or being encoded by nucleic acids contained within the compositions of the present invention (Parmiani, et al. Immunol Lett 2000 Sep 15; 74(1): 41-4; Berzofsky, et al. Nature Immunol. 1: 209-219). Suitable cytokines include, for example, interleukin-2 (IL-2) (Rosenberg, et al. *Nature Med.* 4: 321-327 (1998)), IL-4, IL-7, IL-12 (reviewed by Pardoll, 1992; Harries, et al. J. Gene Med. 2000 Jul-Aug;2(4):243-9; Rao, et al. *J. Immunol.* 156: 3357-3365 (1996)), IL-15 (Xin, et al. *Vaccine*, 17:858-866, 1999), IL-16 (Cruikshank, et al. J. Leuk Biol. 67(6): 757-66, 2000), IL-18 (*J. Cancer Res. Clin. Oncol.* 2001. 127(12): 718-726), GM-CSF (CSF (Disis, et al. *Blood*, 88: 202-210 (1996)), tumor necrosis factor-alpha (TNF-α), or interferons such as IFN-α or INF-γ. Other cytokines may also be suitable for practicing the present invention, as is known in the art.

Chemokines may also be utilized, in either polypeptide or nucleic acid form. Fusion proteins comprising CXCL10 (IP-10) and CCL7 (MCP-3) fused to a tumor self-antigen have been shown to induce anti-tumor immunity (Biragyn, et al. *Nature Biotech.* 1999, 17: 253-258). The chemokines CCL3 (MIP-1α) and CCL5 (RANTES) (Boyer, et al. *Vaccine*, 1999, 17 (Supp.

2): S53-S64) may also be of use in practicing the present invention. Other suitable chemokines are known in the art.

It is also known in the art that suppressive or negative regulatory immune mechanisms may be blocked, resulting in enhanced immune responses. For instance, treatment with anti-CTLA-4 (Shrikant, et al. *Immunity*, 1996, 14: 145-155; Sutmuller, et al. *J. Exp. Med.*, 2001, 194: 823-832), anti-CD25 (Sutmuller, *supra*), anti-CD4 (Matsui, et al. *J. Immunol.*, 1999, 163: 184-193), the fusion protein IL13Ra2-Fc (Terabe, et al. *Nature Immunol.*, 2000, 1: 515-520), and combinations thereof (i.e., anti-CTLA-4 and anti-CD25, Sutmuller, *supra*) have been shown to upregulate anti-tumor immune responses and would be suitable in practicing the present invention. Such treatments, among others, may also be combined with the one or more immunogenic targets of the present invention.

Any of these components may be used alone or in combination with other agents. For instance, it has been shown that a combination of CD80, ICAM-1 and LFA-3 ("TRICOM") may potentiate anti-cancer immune responses (Hodge, et al. *Cancer Res.* 59: 5800-5807 (1999). Other effective combinations include, for example, IL-12 + GM-CSF (Ahlers, et al. *J. Immunol.*, 158: 3947-3958 (1997); Iwasaki, et al. *J. Immunol.* 158: 4591-4601 (1997)), IL-12 + GM-CSF + TNF-α (Ahlers, et al. *Int. Immunol.* 13: 897-908 (2001)), CD80 + IL-12 (Fruend, et al. *Int. J. Cancer*, 85: 508-517 (2000); Rao, et al. *supra*), and CD86 + GM-CSF + IL-12 (Iwasaki, supra). One of skill in the art would be aware of additional combinations useful in carrying out the present invention. In addition, the skilled artisan would be aware of additional reagents or methods that may be used to modulate such mechanisms. These reagents and methods, as well as others known by those of skill in the art, may be utilized in practicing the present invention.

Additional strategies for improving the efficiency of nucleic acid-based immunization may also be used including, for example, the use of self-replicating viral replicons (Caley, et al. 1999. *Vaccine*, 17: 3124-2135; Dubensky, et al. 2000. *Mol. Med.* 6: 723-732; Leitner, et al. 2000. *Cancer Res.* 60: 51-55), codon optimization (Liu, et al. 2000. *Mol. Ther.*, 1: 497-500; Dubensky, *supra*; Huang, et al. 2001. *J. Virol.* 75: 4947-4951), *in vivo* electroporation (Widera, et al. 2000. *J. Immunol.* 164: 4635-3640), incorporation of CpG stimulatory motifs (Gurunathan, et al. *Ann. Rev. Immunol.*, 2000, 18: 927-974; Leitner, *supra*; Cho, et al. J. Immunol. 168(10):4907-13), sequences for targeting of the endocytic or ubiquitin-processing pathways (Thomson, et al. 1998. *J. Virol.* 72: 2246-2252; Velders, et al. 2001. *J. Immunol.*

166: 5366-5373), Marek's disease virus type 1 VP22 sequences (J. Virol. 76(6):2676-82, 2002), prime-boost regimens (Gurunathan, *supra*; Sullivan, et al. 2000. *Nature*, 408: 605-609; Hanke, et al. 1998. *Vaccine*, 16: 439-445; Amara, et al. 2001. *Science*, 292: 69-74), and the use of mucosal delivery vectors such as *Salmonella* (Darji, et al. 1997. *Cell*, 91: 765-775; Woo, et al. 2001. *Vaccine*, 19: 2945-2954). Other methods are known in the art, some of which are described below.

Chemotherapeutic agents, radiation, anti-angiogenic compounds, or other agents may also be utilized in treating and / or preventing cancer using immunogenic targets (Sebti, et al. Oncogene 2000 Dec 27;19(56):6566-73). For example, in treating metastatic melanoma, suitable chemotherapeutic regimens may include BELD (bleomycin, vindesine, lomustine, and deacarbazine; Young, et al. 1985. Cancer, 55: 1879-81), BOLD (bleomycin, vincristine, lomustine, dacarbazine; Seigler, et al. 1980. Cancer, 46 : 2346-8) ; DD (dacarbazine, actinomycin ; Hochster, et al. Cancer Treatment Reports, 69: 39-42), or POC (procarbazine, vincristine, lomustine; Carmo-Pereira, et al. 1984. Cancer Treatment Reports, 68: 1211-4) among others. Other suitable chemotherapeutic regimens may also be utilized.

Many anti-angiogenic agents are known in the art and would be suitable for co-administration with the immunogenic target vaccines and/or chemotherapeutic regimens (see, for example, Timar, et al. 2001. *Pathology Oncol. Res.*, 7(2): 85-94). Such agents include, for example, physiological agents such as growth factors (i.e., ANG-2, NK1,2,4 (HGF), transforming growth factor beta (TGF-β)), cytokines (i.e., interferons such as IFN-α, -β, -γ, platelet factor 4 (PF-4), PR-39), proteases (i.e., cleaved AT-III, collagen XVIII fragment (Endostatin)), HmwKallikrein-d5 plasmin fragment (Angiostatin), prothrombin-F1-2, TSP-1), protease inhibitors (i.e., tissue inhibitor of metalloproteases such as TIMP-1, -2, or –3; maspin; plasminogen activator-inhibitors such as PAI-1; pigment epithelium derived factor (PEDF)), Tumstatin (available through ILEX, Inc.), antibody products (i.e., the collagen-binding antibodies HUIV26, HUI77, XL313; anti-VEGF; anti-integrin (i.e., Vitaxin, (Lxsys))), and glycosidases (i.e., heparinase-I, -III). "Chemical" or modified physiological agents known or believed to have anti-angiogenic potential include, for example, vinblastine, taxol, ketoconazole, thalidomide, dolestatin, combrestatin A, rapamycin (Guba, et al. 2002, *Nature Med.*, 8: 128-135), CEP-7055 (available from Cephalon, Inc.), flavone acetic acid, Bay 12-9566 (Bayer Corp.), AG3340 (Agouron, Inc.), CGS 27023A (Novartis), tetracylcine derivatives (i.e., COL-3

(Collagenix, Inc.)), Neovastat (Aeterna), BMS-275291 (Bristol-Myers Squibb), low dose 5-FU, low dose methotrexate (MTX), irsofladine, radicicol, cyclosporine, captopril, celecoxib, D45152-sulphated polysaccharide, cationic protein (Protamine), cationic peptide-VEGF, Suramin (polysulphonated napthyl urea), compounds that interfere with the function or production of VEGF (i.e., SU5416 or SU6668 (Sugen), PTK787/ZK22584 (Novartis)), Distamycin A, Angiozyme (ribozyme), isoflavinoids, staurosporine derivatives, genistein, EMD121974 (Merck KcgaA), tyrphostins, isoquinolones, retinoic acid, carboxyamidotriazole, TNP-470, octreotide, 2-methoxyestradiol, aminosterols (i.e., squalamine), glutathione analogues (i.e., N-acteyl-L-cysteine), combretastatin A-4 (Oxigene), Eph receptor blocking agents (*Nature*, 414:933-938, 2001), Rh-Angiostatin, Rh-Endostatin (WO 01/93897), cyclic-RGD peptide, accutin-disintegrin, benzodiazepenes, humanized anti-avb3 Ab, Rh-PAI-2, amiloride, p-amidobenzamidine, anti-uPA ab, anti-uPAR Ab, L-phanylalanin-N-methylamides (i.e., Batimistat, Marimastat), AG3340, and minocycline. Many other suitable agents are known in the art and would suffice in practicing the present invention.

The present invention may also be utilized in combination with "non-traditional" methods of treating cancer. For example, it has recently been demonstrated that administration of certain anaerobic bacteria may assist in slowing tumor growth. In one study, *Clostridium novyi* was modified to eliminate a toxin gene carried on a phage episome and administered to mice with colorectal tumors (Dang, et al. *P.N.A.S. USA*, 98(26): 15155-15160, 2001). In combination with chemotherapy, the treatment was shown to cause tumor necrosis in the animals. The reagents and methodologies described in this application may be combined with such treatment methodologies.

Nucleic acids encoding immunogenic targets may be administered to patients by any of several available techniques. Various viral vectors that have been successfully utilized for introducing a nucleic acid to a host include retrovirus, adenovirus, adeno-associated virus (AAV), herpes virus, and poxvirus, among others. It is understood in the art that many such viral vectors are available in the art. The vectors of the present invention may be constructed using standard recombinant techniques widely available to one skilled in the art. Such techniques may be found in common molecular biology references such as *Molecular Cloning: A Laboratory Manual* (Sambrook, et al., 1989, Cold Spring Harbor Laboratory Press), *Gene Expression Technology* (Methods in Enzymology, Vol. 185, edited by D. Goeddel, 1991. Academic Press,

San Diego, CA), and *PCR Protocols: A Guide to Methods and Applications* (Innis, et al. 1990. Academic Press, San Diego, CA).

Preferred retroviral vectors are derivatives of lentivirus as well as derivatives of murine or avian retroviruses. Examples of suitable retroviral vectors include, for example, Moloney murine leukemia virus (MoMuLV), Harvey murine sarcoma virus (HaMuSV), murine mammary tumor virus (MuMTV), SIV, BIV, HIV and Rous Sarcoma Virus (RSV). A number of retroviral vectors can incorporate multiple exogenous nucleic acid sequences. As recombinant retroviruses are defective, they require assistance in order to produce infectious vector particles. This assistance can be provided by, for example, helper cell lines encoding retrovirus structural genes. Suitable helper cell lines include Ψ2, PA317 and PA12, among others. The vector virions produced using such cell lines may then be used to infect a tissue cell line, such as NIH 3T3 cells, to produce large quantities of chimeric retroviral virions. Retroviral vectors may be administered by traditional methods (i.e., injection) or by implantation of a "producer cell line" in proximity to the target cell population (Culver, K., *et al.*, 1994, *Hum. Gene Ther.*, 5 (3): 343-79; Culver, K., *et al.*, *Cold Spring Harb. Symp. Quant. Biol.*, 59: 685-90); Oldfield, E., 1993, *Hum. Gene Ther.*, 4 (1): 39-69). The producer cell line is engineered to produce a viral vector and releases viral particles in the vicinity of the target cell. A portion of the released viral particles contact the target cells and infect those cells, thus delivering a nucleic acid of the present invention to the target cell. Following infection of the target cell, expression of the nucleic acid of the vector occurs.

Adenoviral vectors have proven especially useful for gene transfer into eukaryotic cells (Rosenfeld, M., *et al.*, 1991, *Science*, 252 (5004): 431-4; Crystal, R., *et al.*, 1994, *Nat. Genet.*, 8 (1): 42-51), the study eukaryotic gene expression (Levrero, M., *et al.*, 1991, *Gene*, 101 (2): 195-202), vaccine development (Graham, F. and Prevec, L., 1992, *Biotechnology*, 20: 363-90), and in animal models (Stratford-Perricaudet, L., *et al.*, 1992, *Bone Marrow Transplant.*, 9 (Suppl. 1): 151-2 ; Rich, D., *et al.*, 1993, *Hum. Gene Ther.*, 4 (4): 461-76). Experimental routes for administrating recombinant Ad to different tissues *in vivo* have included intratracheal instillation (Rosenfeld, M., *et al.*, 1992, *Cell*, 68 (1): 143-55) injection into muscle (Quantin, B., *et al.*, 1992, *Proc. Natl. Acad. Sci. U.S.A.*, 89 (7): 2581-4), peripheral intravenous injection (Herz, J., and Gerard, R., 1993, *Proc. Natl. Acad. Sci. U.S.A.*, 90 (7): 2812-6) and stereotactic inoculation to brain (Le Gal La Salle, G., *et al.*, 1993, *Science*, 259 (5097): 988-90), among others.

Adeno-associated virus (AAV) demonstrates high-level infectivity, broad host range and specificity in integrating into the host cell genome (Hermonat, P., et al., 1984, *Proc. Natl. Acad. Sci. U.S.A.*, 81 (20): 6466-70). And Herpes Simplex Virus type-1 (HSV-1) is yet another attractive vector system, especially for use in the nervous system because of its neurotropic property (Geller, A., *et al.*, 1991, *Trends Neurosci.*, 14 (10): 428-32; Glorioso, *et al.*, 1995, *Mol. Biotechnol.*, 4 (1): 87-99; Glorioso, *et al.*, 1995, *Annu. Rev. Microbiol.*, 49: 675-710).

Poxvirus is another useful expression vector (Smith, et al. 1983, *Gene*, 25 (1): 21-8; Moss, et al, 1992, *Biotechnology*, 20: 345-62; Moss, et al, 1992, *Curr. Top. Microbiol. Immunol.*, 158: 25-38; Moss, et al. 1991. *Science*, 252: 1662-1667). Poxviruses shown to be useful include vaccinia, NYVAC, avipox, fowlpox, canarypox, ALVAC, and ALVAC(2), among others.

NYVAC (vP866) was derived from the Copenhagen vaccine strain of vaccinia virus by deleting six nonessential regions of the genome encoding known or potential virulence factors (see, for example, U.S. Pat. Nos. 5,364,773 and 5,494,807). The deletion loci were also engineered as recipient loci for the insertion of foreign genes. The deleted regions are: thymidine kinase gene (TK; J2R); hemorrhagic region (u; B13R+B14R); A type inclusion body region (ATI; A26L); hemagglutinin gene (HA; A56R); host range gene region (C7L-K1L); and, large subunit, ribonucleotide reductase (I4L). NYVAC is a genetically engineered vaccinia virus strain that was generated by the specific deletion of eighteen open reading frames encoding gene products associated with virulence and host range. NYVAC has been show to be useful for expressing TAs (see, for example, U.S. Pat. No. 6,265,189). NYVAC (vP866), vP994, vCP205, vCP1433, placZH6H4Lreverse, pMPC6H6K3E3 and pC3H6FHVB were also deposited with the ATCC under the terms of the Budapest Treaty, accession numbers VR-2559, VR-2558, VR-2557, VR-2556, ATCC-97913, ATCC-97912, and ATCC-97914, respectively.

ALVAC-based recombinant viruses (i.e., ALVAC-1 and ALVAC-2) are also suitable for use in practicing the present invention (see, for example, U.S. Pat. No. 5,756,103). ALVAC(2) is identical to ALVAC(1) except that ALVAC(2) genome comprises the vaccinia E3L and K3L genes under the control of vaccinia promoters (U.S. Pat. No. 6,130,066; Beattie et al., 1995a, 1995b, 1991; Chang et al., 1992; Davies et al., 1993). Both ALVAC(1) and ALVAC(2) have been demonstrated to be useful in expressing foreign DNA sequences, such as TAs (Tartaglia et al., 1993 a,b; U.S. Pat. No. 5,833,975). ALVAC was deposited under the terms of the Budapest

Treaty with the American Type Culture Collection (ATCC), 10801 University Boulevard, Manassas, Va. 20110-2209, USA, ATCC accession number VR-2547.

Another useful poxvirus vector is TROVAC. TROVAC refers to an attenuated fowlpox that was a plaque-cloned isolate derived from the FP-1 vaccine strain of fowlpoxvirus which is licensed for vaccination of 1 day old chicks. TROVAC was likewise deposited under the terms of the Budapest Treaty with the ATCC, accession number 2553.

"Non-viral" plasmid vectors may also be suitable in practicing the present invention. Preferred plasmid vectors are compatible with bacterial, insect, and / or mammalian host cells. Such vectors include, for example, PCR-II, pCR3, and pcDNA3.1 (Invitrogen, San Diego, CA), pBSII (Stratagene, La Jolla, CA), pET15 (Novagen, Madison, WI), pGEX (Pharmacia Biotech, Piscataway, NJ), pEGFP-N2 (Clontech, Palo Alto, CA), pETL (BlueBacII, Invitrogen), pDSR-alpha (PCT pub. No. WO 90/14363) and pFastBacDual (Gibco-BRL, Grand Island, NY) as well as Bluescript® plasmid derivatives (a high copy number COLE1-based phagemid, Stratagene Cloning Systems, La Jolla, CA), PCR cloning plasmids designed for cloning Taq-amplified PCR products (*e.g.*, TOPO™ TA cloning® kit, PCR2.1® plasmid derivatives, Invitrogen, Carlsbad, CA). Bacterial vectors may also be used with the current invention. These vectors include, for example, *Shigella, Salmonella, Vibrio cholerae, Lactobacillus, Bacille calmette guérin (BCG), and Streptococcus* (see for example, WO 88/6626; WO 90/0594; WO 91/13157; WO 92/1796; and WO 92/21376). Many other non-viral plasmid expression vectors and systems are known in the art and could be used with the current invention.

Suitable nucleic acid delivery techniques include DNA-ligand complexes, adenovirus-ligand-DNA complexes, direct injection of DNA, $CaPO_4$ precipitation, gene gun techniques, electroporation, and colloidal dispersion systems, among others. Colloidal dispersion systems include macromolecule complexes, nanocapsules, microspheres, beads, and lipid-based systems including oil-in-water emulsions, micelles, mixed micelles, and liposomes. The preferred colloidal system of this invention is a liposome, which are artificial membrane vesicles useful as delivery vehicles *in vitro* and *in vivo*. RNA, DNA and intact virions can be encapsulated within the aqueous interior and be delivered to cells in a biologically active form (Fraley, R., *et al.*, 1981, *Trends Biochem. Sci.*, 6: 77). The composition of the liposome is usually a combination of phospholipids, particularly high-phase-transition-temperature phospholipids, usually in

combination with steroids, especially cholesterol. Other phospholipids or other lipids may also be used. The physical characteristics of liposomes depend on pH, ionic strength, and the presence of divalent cations. Examples of lipids useful in liposome production include phosphatidyl compounds, such as phosphatidylglycerol, phosphatidylcholine, phosphatidylserine, phosphatidylethanolamine, sphingolipids, cerebrosides, and gangliosides. Particularly useful are diacylphosphatidylglycerols, where the lipid moiety contains from 14-18 carbon atoms, particularly from 16-18 carbon atoms, and is saturated. Illustrative phospholipids include egg phosphatidylcholine, dipalmitoylphosphatidylcholine and distearoylphosphatidylcholine.

An immunogenic target may also be administered in combination with one or more adjuvants to boost the immune response. Exemplary adjuvants are shown in Table II below:

Table II

Types of Immunologic Adjuvants

Type of Adjuvant	General Examples	Specific Examples/References
Gel-type	Aluminum hydroxide/phosphate ("alum adjuvants")	(Aggerbeck and Heron, 1995)
	Calcium phosphate	(Relyveld, 1986)
Microbial	Muramyl dipeptide (MDP)	(Chedid et al., 1986)
	Bacterial exotoxins	Cholera toxin (CT), *E.coli* labile toxin (LT)(Freytag and Clements, 1999)
	Endotoxin-based adjuvants	Monophosphoryl lipid A (MPL) (Ulrich and Myers, 1995)
	Other bacterial	CpG oligonucleotides (Corral and Petray, 2000), BCG sequences (Krieg, et al. *Nature*, 374:576), tetanus toxoid (Rice, et al. *J. Immunol.*, 2001, 167: 1558-1565)
Particulate	Biodegradable Polymer microspheres	(Gupta et al., 1998)
	Immunostimulatory complexes (ISCOMs)	(Morein and Bengtsson, 1999)
	Liposomes	(Wassef et al., 1994)
Oil-emulsion and surfactant-based adjuvants	Freund's incomplete adjuvant	(Jensen et al., 1998)
	Microfluidized emulsions	MF59 (Ott et al., 1995)
		SAF (Allison and Byars, 1992) (Allison, 1999)
	Saponins	QS-21 (Kensil, 1996)
Synthetic	Muramyl peptide derivatives	Murabutide (Lederer, 1986) Threony-MDP (Allison, 1997)
	Nonionic block copolymers	L121 (Allison, 1999)
	Polyphosphazene (PCPP)	(Payne et al., 1995)

	Synthetic polynucleotides	Poly A:U, Poly I:C (Johnson, 1994)
	Thalidomide derivatives	CC-4047/ACTIMID (J. Immunol., 168(10):4914-9)

Administration of a composition of the present invention to a host may be accomplished using any of a variety of techniques known to those of skill in the art. The composition(s) may be processed in accordance with conventional methods of pharmacy to produce medicinal agents for administration to patients, including humans and other mammals (i.e., a "pharmaceutical composition"). The pharmaceutical composition is preferably made in the form of a dosage unit containing a given amount of DNA, viral vector particles, polypeptide or peptide, for example. A suitable daily dose for a human or other mammal may vary widely depending on the condition of the patient and other factors, but, once again, can be determined using routine methods.

The pharmaceutical composition may be administered orally, parentally, by inhalation spray, rectally, intranodally, or topically in dosage unit formulations containing conventional pharmaceutically acceptable carriers, adjuvants, and vehicles. The term "pharmaceutically acceptable carrier" or "physiologically acceptable carrier" as used herein refers to one or more formulation materials suitable for accomplishing or enhancing the delivery of a nucleic acid, polypeptide, or peptide as a pharmaceutical composition. A "pharmaceutical composition" is a composition comprising a therapeutically effective amount of a nucleic acid or polypeptide. The terms "effective amount" and "therapeutically effective amount" each refer to the amount of a nucleic acid or polypeptide used to induce or enhance an effective immune response. It is preferred that compositions of the present invention provide for the induction or enhancement of an anti-tumor immune response in a host which protects the host from the development of a tumor and / or allows the host to eliminate an existing tumor from the body.

For oral administration, the pharmaceutical composition may be of any of several forms including, for example, a capsule, a tablet, a suspension, or liquid, among others. Liquids may be administered by injection as a composition with suitable carriers including saline, dextrose, or water. The term parenteral as used herein includes subcutaneous, intravenous, intramuscular, intrasternal, infusion, or intraperitoneal administration. Suppositories for rectal administration of the drug can be prepared by mixing the drug with a suitable non-irritating excipient such as cocoa butter and polyethylene glycols that are solid at ordinary temperatures but liquid at the rectal temperature.

The dosage regimen for immunizing a host or otherwise treating a disorder or a disease with a composition of this invention is based on a variety of factors, including the type of disease, the age, weight, sex, medical condition of the patient, the severity of the condition, the route of administration, and the particular compound employed. For example, a poxviral vector may be administered as a composition comprising 1 x 10^6 infectious particles per dose. Thus, the dosage regimen may vary widely, but can be determined routinely using standard methods.

A prime-boost regimen may also be utilized (WO 01/30382 A1) in which the targeted immunogen is initially administered in a priming step in one form followed by a boosting step in which the targeted immunogen is administered in another form. The form of the targeted immunogen in the priming and boosting steps are different. For instance, if the priming step utilized a nucleic acid, the boost may be administered as a peptide. Similarly, where a priming step utilized one type of recombinant virus (i.e., ALVAC), the boost step may utilize another type of virus (i.e., NYVAC). This prime-boost method of administration has been shown to induce strong immunological responses. Various combinations of forms are suitable in practicing the present invention.

While the compositions of the invention can be administered as the sole active pharmaceutical agent, they can also be used in combination with one or more other compositions or agents (i.e., other immunogenic targets, co-stimulatory molecules, adjuvants). When administered as a combination, the individual components can be formulated as separate compositions administered at the same time or different times, or the components can be combined as a single composition.

Injectable preparations, such as sterile injectable aqueous or oleaginous suspensions, may be formulated according to known methods using suitable dispersing or wetting agents and suspending agents. The injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally acceptable diluent or solvent. Suitable vehicles and solvents that may be employed are water, Ringer's solution, and isotonic sodium chloride solution, among others. For instance, a viral vector such as a poxvirus may be prepared in 0.4% NaCl. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose, any bland fixed oil may be employed, including synthetic mono- or diglycerides. In addition, fatty acids such as oleic acid find use in the preparation of injectables.

For topical administration, a suitable topical dose of a composition may be administered one to four, and preferably two or three times daily. The dose may also be administered with intervening days during which no does is applied. Suitable compositions may comprise from 0.001% to 10% w/w, for example, from 1% to 2% by weight of the formulation, although it may comprise as much as 10% w/w, but preferably not more than 5% w/w, and more preferably from 0.1% to 1% of the formulation. Formulations suitable for topical administration include liquid or semi-liquid preparations suitable for penetration through the skin (*e.g.*, liniments, lotions, ointments, creams, or pastes) and drops suitable for administration to the eye, ear, or nose.

The pharmaceutical compositions may also be prepared in a solid form (including granules, powders or suppositories). The pharmaceutical compositions may be subjected to conventional pharmaceutical operations such as sterilization and/or may contain conventional adjuvants, such as preservatives, stabilizers, wetting agents, emulsifiers, buffers etc. Solid dosage forms for oral administration may include capsules, tablets, pills, powders, and granules. In such solid dosage forms, the active compound may be admixed with at least one inert diluent such as sucrose, lactose, or starch. Such dosage forms may also comprise, as in normal practice, additional substances other than inert diluents, *e.g.*, lubricating agents such as magnesium stearate. In the case of capsules, tablets, and pills, the dosage forms may also comprise buffering agents. Tablets and pills can additionally be prepared with enteric coatings. Liquid dosage forms for oral administration may include pharmaceutically acceptable emulsions, solutions, suspensions, syrups, and elixirs containing inert diluents commonly used in the art, such as water. Such compositions may also comprise adjuvants, such as wetting sweetening, flavoring, and perfuming agents.

Pharmaceutical compositions comprising a nucleic acid or polypeptide of the present invention may take any of several forms and may be administered by any of several routes. In preferred embodiments, the compositions are administered via a parenteral route (intradermal, intramuscular or subcutaneous) to induce an immune response in the host. Alternatively, the composition may be administered directly into a lymph node (intranodal) or tumor mass (i.e., intratumoral administration). For example, the dose could be administered subcutaneously at days 0, 7, and 14. Suitable methods for immunization using compositions comprising TAs are known in the art, as shown for p53 (Hollstein et al., 1991), p21-ras (Almoguera et al., 1988), HER-2 (Fendly et al., 1990), the melanoma-associated antigens (MAGE-1; MAGE-2) (van der

Bruggen et al., 1991), p97 (Hu et al., 1988), melanoma-associated antigen E (WO 99/30737) and carcinoembryonic antigen (CEA) (Kantor et al., 1993; Fishbein et al., 1992; Kaufman et al., 1991), among others.

Preferred embodiments of administratable compositions include, for example, nucleic acids or polypeptides in liquid preparations such as suspensions, syrups, or elixirs. Preferred injectable preparations include, for example, nucleic acids or polypeptides suitable for parental, subcutaneous, intradermal, intramuscular or intravenous administration such as sterile suspensions or emulsions. For example, a recombinant poxvirus may be in admixture with a suitable carrier, diluent, or excipient such as sterile water, physiological saline, glucose or the like. The composition may also be provided in lyophilized form for reconstituting, for instance, in isotonic aqueous, saline buffer. In addition, the compositions can be co-administered or sequentially administered with other antineoplastic, anti-tumor or anti-cancer agents and/or with agents which reduce or alleviate ill effects of antineoplastic, anti-tumor or anti-cancer agents.

A kit comprising a composition of the present invention is also provided. The kit can include a separate container containing a suitable carrier, diluent or excipient. The kit can also include an additional anti-cancer, anti-tumor or antineoplastic agent and/or an agent that reduces or alleviates ill effects of antineoplastic, anti-tumor or anti-cancer agents for co- or sequential-administration. Additionally, the kit can include instructions for mixing or combining ingredients and/or administration.

A better understanding of the present invention and of its many advantages will be had from the following examples, given by way of illustration.

EXAMPLES

Example 1

Construction of the Multi-Antigen Construct vT416

The expression vector vT416 (ALVAC-NY-ESO-1/Trp-2-LFA-3/ICAM-1/B7.1-E3L/K3L) was constructed in the ALVAC vector using standard techniques. DNA sequences encoding NY-ESO-1, Trp-2, LFA-3, ICAM-1, B7.1, vvE3L and vvK3L were inserted into various loci within the ALVAC genome. DNA sequences encoding NY-ESO-1 (Chen et al. 1997 PNAS 94:1914) and TRP-2 (Wang et al. 1996 J. Exp. Med. 184:2207) were inserted into

the C5 locus. DNA sequences encoding LFA-3 (Wallner, et al. (1987) J. Exp. Med. 166:923-932), ICAM-1 (Staunton, et al. (1988) Cell 52:925-933) and B7.1 (Chen, et al. (1992) Cell 71:1093-1102) were inserted into the C3 locus. LFA-3, ICAM-1 and B7.1 form an expression cassette known as TRICOM. DNA sequences encoding vvE3L (Chang, et al. 1992. Proc. Natl. Acad. Sci. U. S. A 89:4825-4829) and vvK3L (Beattie, et al. 1991. Virology 183:419-422) were inserted into the C6 locus. Promoters were utilized as follows:

Table III

DNA sequence	Promoter
E3L	vaccinia E3L
K3L	vaccinia H6
LFA-3	vaccinia 30K
ICAM-1	vaccinia I3
B7.1	sE/L
NY-ESO-1	vaccinia H6
TRP-2	sE/L

Promoter sE/L is described by Chakrabarti, et al. (BioTechniques 23: 1094-1097, 1997).

The donor plasmids utilized are shown below:

Table IV

Plasmid	Size (bp)	Vector	Antibiotic Resitance Gene
pMPC6H6K3E3	-	pBS-SK	Amp
pALVAC.Tricom(C3) #33	10,470	pBS-SK	Amp
pT1132	11,154	pBS-SK	Amp

NY-ESO-1 and TRP-2 DNA sequences were inserted into the ALVAC donor plasmid pT1132. This donor plasmid was then used with pALVAC.Tricom(C3) #33 to generate the ALVAC-TRICOM recombinant expressing these genes using standard techniques. The plasmids pALVAC.Tricom(C3) #33 and pT1132 are shown in **Figure 1**. The DNA sequences of pALVAC.Tricom(C3) #33 and pT1132 are shown in **Figures 2** and **3**, respectively.

Example 2

Construction of the Multi-Antigen Construct vT419

The expression vector vT419 (ALVAC-gp100M/Mart-1/ Mage-1,3 minigene-LFA-3/ICAM-1/B7.1-E3L/K3L) was constructed in the ALVAC vector using standard techniques. DNA sequences encoding the gp100M/MART-1/MAGE-1,3 minigene, LFA-3, ICAM-1, B7.1, vvE3L and vvK3L were inserted into various loci within the ALVAC genome. The gp100M/ MART-1/MAGE-1,3 minigene was inserted into the C5 locus. DNA sequences encoding LFA-3 (Wallner, et al. (1987) J. Exp. Med. 166:923-932), ICAM-1 (Staunton, et al. (1988) Cell 52:925-933) and B7.1 (Chen, et al. (1992) Cell 71:1093-1102) were inserted into the C3 locus. LFA-3, ICAM-1 and B7.1 form an expression cassette known as TRICOM. DNA sequences encoding vvE3L (Chang, et al. 1992. Proc. Natl. Acad. Sci. U. S. A 89:4825-4829) and vvK3L (Beattie, et al. 1991. Virology 183:419-422) were inserted into the C6 locus. Promoters were utilized as follows:

Table V

Gene	Promoter
E3L	vaccinia E3L
K3L	vaccinia H6
LFA-3	vaccinia 30K
ICAM-1	vaccinia I3
B7.1	sE/L
gp100(M)	vaccinia H6
Mart-1	vaccinia 42K

Promoter sE/L is described by Chakrabarti, et al. (BioTechniques 23: 1094-1097, 1997).

The donor plasmids utilized are shown below:

Table VI

Plasmid	Size (bp)	Vector	Antibiotic Resitance Gene
PMPC6H6K3E3	-	pBS-SK	Amp
pALVAC.Tricom(C3) #33	10,470	pBS-SK	Amp
pT3217	11,465	pBS-SK	Amp

gp100(M), Mart-1 and Mage-1,3 minigene were inserted into the ALVAC C5 donor plasmid pT3217. This donor plasmid was then used with pALVAC.Tricom(C3) #33 to generate the ALVAC-TRICOM recombinant expressing these genes using standard techniques. This donor plasmid inserts into the C5 site. pALVAC.Tricom(C3) #33 is shown in **Figures 1** and **2.** The pT3217 plasmid is shown in **Figure 4.** The DNA sequence of pT3217 is shown in **Figure 5.**

EXAMPLE 3

Immunological Assessment of Multi-Antigen Vectors

The results of the first animal experiment indicated a trend toward higher immunological responses to three (Mart 1, NY-ESO-1 and gp100) of the four antigens when the vaccine was given as two separate injections. However, these differences were not statistically significant. In detail, HLA-A2/K^b transgenic mice (5/group) were immunized subcutaneously with vT419 (ALVAC(2)-gp100M/MART-1/MAGE-1/3 minigene/TRICOM) and vT416 (ALVAC(2)-TRP-2/NY-ESO-1/TRICOM) either combined at one site or given as separate injections. Control mice were immunized with parental ALVAC(2). Mice were vaccinated three times (at three week intervals), and three weeks after the last boost T cell responses in individual mice were analyzed by IFN-g ELISPOT and CTL assays following in vitro restimulation with peptide. Compared to control animals, mice vaccinated with the multi-antigen vectors (at 2 sites) exhibited statistically significant ELISPOT responses against MART-1. The IFN-gamma response to gp100M and NY-ESO-1 were also detectable, although these responses were not statistically significant due to response variability and the small number of cultures tested. ELISPOT responses against the TRP-2 antigen were elevated in all groups tested (including control animals), presumably due to the fact that the dominant A2-restricted TRP-2 peptide (180-188) cross-reacts with H-$2K^b$ and can induce low avidity T cell responses in naïve mice following in vitro culture, and were therefore not statistically significant. Interestingly, ELISPOT responses in mice injected with an admixture of vT416 and vT419 were generally lower than in mice receiving each virus separately, although these differences did not achieve statistical significance. The CTL data were largely negative, except for one strong anti-gp100 response and one marginal anti-MART-1 response, both of which occurred in mice vaccinated with vT416 and vT419 (two sites). Overall, these results provided encouraging data that establish that the multi-antigen vectors can generate

responses against MART-1, and suggest that anti-gp100 and anti-NY-ESO-1 responses can also be induced.

Two additional pre-clinical animal studies have been completed using the melanoma multi-antigen ALVAC recombinants. In these experiments, HLA-A2/K^b transgenic mice (5/group) were immunized subcutaneously with vT419 (ALVAC(2)-gp100M/MART-1/MAGE-1/3 minigene/TRICOM) and vT416 (ALVAC(2)-TRP-2/NY-ESO-1/TRICOM) either combined at one site or given as separate injections. Control mice were immunized with parental ALVAC(2). After vaccination, the T cell responses in individual mice were assessed by IFN-gamma ELISPOT assay following in vitro restimulation with peptide. Unlike the previous multi-antigen experiment, which provided encouraging immunogenicity data, the two most recent studies generated inconclusive data, due to high background responses in control immunized animals. Therefore, overall the results were deemed as inconclusive.

To confirm the immunogenicity of the multi-antigen constructs, and to repeat results from the first study, another pre-clinical animal study has been completed. HLA-A2/K^b transgenic mice (10/group) were immunized subcutaneously with vT419 (ALVAC(2)-gp100M/MART-1/MAGE-1/3 minigene/TRICOM) and vT416 (ALVAC(2)-TRP-2/NY-ESO-1/TRICOM) given as separate injections. Control mice were immunized with parental ALVAC(2). Statistically significant ELISPOT responses were detectable against gp100, Mart-1 and TRP-2, and some responses were detected against NY-ESO-1, which were at the border of being statistically significant.

While the present invention has been described in terms of the preferred embodiments, it is understood that variations and modifications will occur to those skilled in the art. Therefore, it is intended that the appended claims cover all such equivalent variations that come within the scope of the invention as claimed.

CLAIMS

What is claimed is:

1. An expression vector for co-expressing at least two immunogenic targets, wherein said immunogenic targets are selected from the group consisting of NY-ESO-1, TRP-2, gp100, gp100M, a MART antigen, MART-1, a MAGE antigen, MAGE-1, and MAGE-3.
2. The expression vector of claim 1 wherein the vector is a plasmid or a viral vector.
3. The expression vector of claim 2 wherein the viral vector is selected from the group consisting of poxvirus, adenovirus, retrovirus, herpesvirus, and adeno-associated virus.
4. The expression vector of claim 3 wherein the viral vector is a poxvirus selected from the group consisting of vaccinia, NYVAC, avipox, canarypox, ALVAC, ALVAC(2), fowlpox, and TROVAC.
5. The expression vector of claim 4 wherein the viral vector is a poxvirus selected from the group consisting of NYVAC, ALVAC, and ALVAC(2).
6. The expression vector of claim 1 further comprising at least one nucleic sequence encoding an angiogenesis-associated antigen.
7. The expression vector of claim 6 wherein the vector is a plasmid or a viral vector.
8. The expression vector of claim 7 wherein the viral vector is selected from the group consisting of poxvirus, adenovirus, retrovirus, herpesvirus, and adeno-associated virus.
9. The expression vector of claim 8 wherein the viral vector is a poxvirus selected from the group consisting of vaccinia, NYVAC, avipox, canarypox, ALVAC, ALVAC(2), fowlpox, and TROVAC.
10. The expression vector of claim 9 wherein the viral vector is a poxvirus selected from the group consisting of NYVAC, ALVAC, and ALVAC(2).
11. The expression vector of claim 1 or 6 further comprising at least one nucleic acid sequence encoding a co-stimulatory component.
12. The expression vector of claim 11 wherein the vector is a plasmid or a viral vector.
13. The expression vector of claim 12 wherein the viral vector is selected from the group consisting of poxvirus, adenovirus, retrovirus, herpesvirus, and adeno-associated virus.
14. The expression vector of claim 13 wherein the viral vector is a poxvirus selected from the group consisting of vaccinia, NYVAC, avipox, canarypox, ALVAC, ALVAC(2), fowlpox, and TROVAC.

15. The expression vector of claim 14 wherein the viral vector is a poxvirus selected from the group consisting of NYVAC, ALVAC, and ALVAC(2).

16. The expression vector of any one claims 11-15 wherein the co-stimulatory component is human B7.1.

17. A composition comprising an expression vector of any one of claims 1-16 in a pharmaceutically acceptable carrier.

18. A method for preventing or treating cancer comprising administering to a host an expression vector of any one of claims 1-16.

19. A method for preventing or treating cancer comprising administering to a host a composition of claim 17.

FIGURE 1

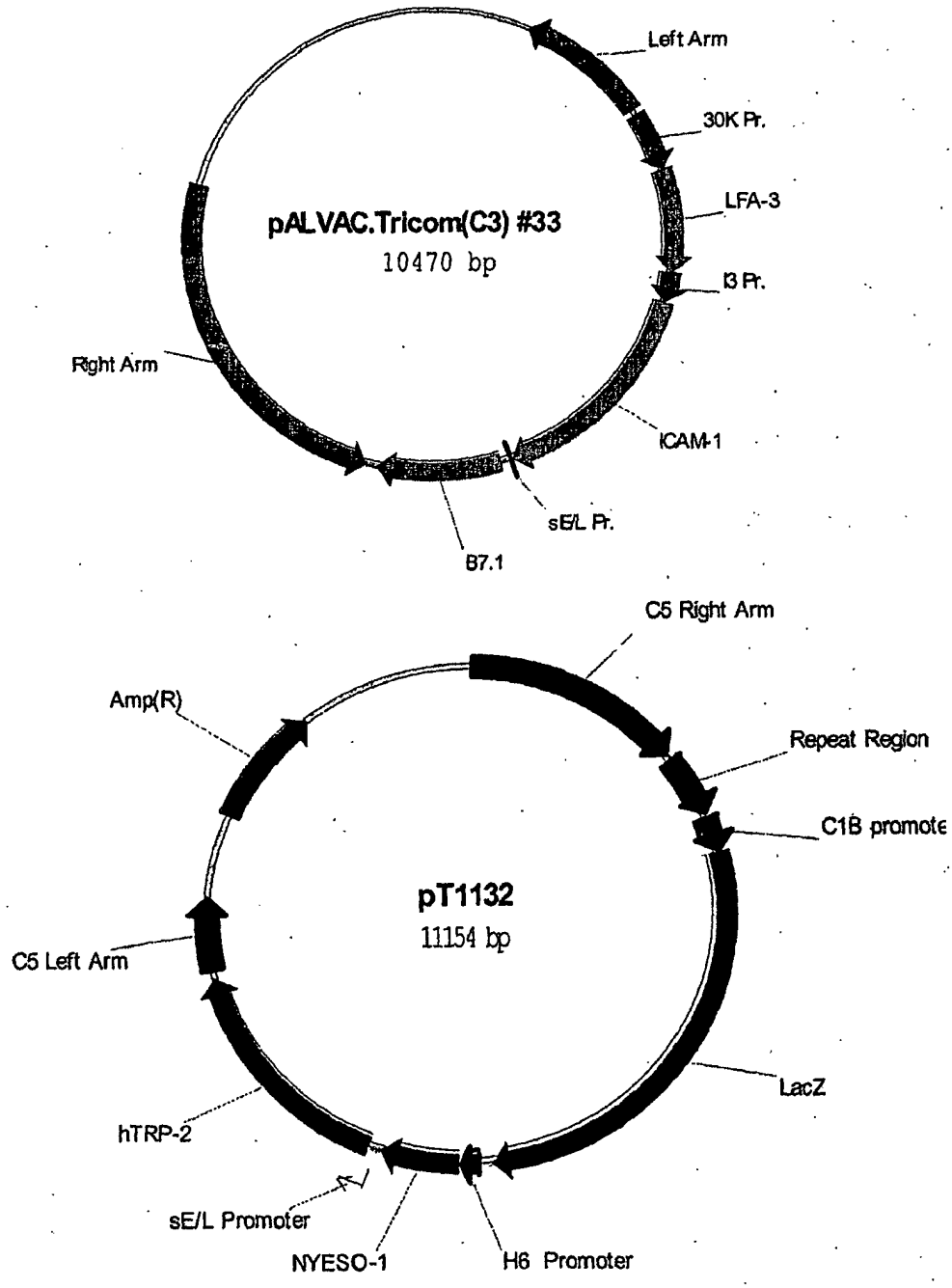

Left Arm
30K Pr.
LFA-3
I3 Pr.
ICAM-1
sE/L Pr.
B7.1
Right Arm
pALVAC.Tricom(C3) #33
10470 bp
C5 Right Arm
Repeat Region
C1B promote
LacZ
H6 Promoter
NYESO-1
sE/L Promoter
hTRP-2
C5 Left Arm
Amp(R)
pT1132
11154 bp

FIGURE 2

DNA Sequence of pALVAC.Tricom(C3) #33

```
     1  GGAAATTGTA AACGTTAATA TTTTGTTAAA ATTCGCGTTA AATTTTTGTT
        CCTTTAACAT TTGCAATTAT AAAACAATTT TAAGCGCAAT TTAAAAACAA
    51  AAATCAGCTC ATTTTTTAAC CAATAGGCCG AAATCGGCAA AATCCCTTAT
        TTTAGTCGAG TAAAAAATTG GTTATCCGGC TTTAGCCGTT TTAGGGAATA
   101  AAATCAAAAG AATAGACCGA GATAGGGTTG AGTGTTGTTC CAGTTTGGAA
        TTTAGTTTTC TTATCTGGCT CTATCCCAAC TCACAACAAG GTCAAACCTT
   151  CAAGAGTCCA CTATTAAAGA ACGTGGACTC CAACGTCAAA GGGCGAAAAA
        GTTCTCAGGT GATAATTTCT TGCACCTGAG GTTGCAGTTT CCCGCTTTTT
   201  CCGTCTATCA GGGCGATGGC CCACTACGTG AACCATCACC CTAATCAAGT
        GGCAGATAGT CCCGCTACCG GGTGATGCAC TTGGTAGTGG GATTAGTTCA
   251  TTTTTGGGGT CGAGGTGCCG TAAAGCACTA AATCGGAACC CTAAAGGGAG
        AAAAACCCCA GCTCCACGGC ATTTCGTGAT TTAGCCTTGG GATTTCCCTC
   301  CCCCCGATTT AGAGCTTGAC GGGGAAAGCC GGCGAACGTG GCGAGAAAGG
        GGGGGCTAAA TCTCGAACTG CCCCTTTCGG CCGCTTGCAC CGCTCTTTCC
   351  AAGGGAAGAA AGCGAAAGGA GCGGGCGCTA GGGCGCTGGC AAGTGTAGCG
        TTCCCTTCTT TCGCTTTCCT CGCCCGCGAT CCCGCGACCG TTCACATCGC
   401  GTCACGCTGC GCGTAACCAC CACACCCGCC GCGCTTAATG CGCCGCTACA
        CAGTGCGACG CGCATTGGTG GTGTGGGCGG CGCGAATTAC GCGGCGATGT
   451  GGGCGCGTCG CGCCATTCGC CATTCAGGCT GCGCAACTGT TGGGAAGGGC
        CCCGCGCAGC GCGGTAAGCG GTAAGTCCGA CGCGTTGACA ACCCTTCCCG
   501  GATCGGTGCG GGCCTCTTCG CTATTACGCC AGCTGGCGAA AGGGGGATGT
        CTAGCCACGC CCGGAGAAGC GATAATGCGG TCGACCGCTT TCCCCCTACA
   551  GCTGCAAGGC GATTAAGTTG GGTAACGCCA GGGTTTTCCC AGTCACGACG
        CGACGTTCCG CTAATTCAAC CCATTGCGGT CCCAAAAGGG TCAGTGCTGC
   601  TTGTAAAACG ACGGCCAGTG AATTGTAATA CGACTCACTA TAGGGCGAAT
        AACATTTTGC TGCCGGTCAC TTAACATTAT GCTGAGTGAT ATCCCGCTTA
   651  TGGGTACCGC GGCCGCGTCG ACATGCATTG TTAGTTCTGT AGATCAGTAA
        ACCCATGGCG CCGGCGCAGC TGTACGTAAC AATCAAGACA TCTAGTCATT
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
   701  CGTATAGCAT ACGAGTATAA TTATCGTAGG TAGTAGGTAT CCTAAAATAA
        GCATATCGTA TGCTCATATT AATAGCATCC ATCATCCATA GGATTTTATT
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                              Left Arm
   751  ATCTGATACA GATAATAACT TTGTAAATCA ATTCAGCAAT TTCTCTATTA
        TAGACTATGT CTATTATTGA AACATTTAGT TAAGTCGTTA AAGAGATAAT
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                              Left Arm
   801  TCATGATAAT GATTAATACA CAGCGTGTCG TTATTTTTTG TTACGATAGT
        AGTACTATTA CTAATTATGT GTCGCACAGC AATAAAAAAC AATGCTATCA
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                              Left Arm
   851  ATTTCTAAAG TAAAGAGCAG GAATCCCTAG TATAATAGAA ATAATCCATA
        TAAAGATTTC ATTTCTCGTC CTTAGGGATC ATATTATCTT TATTAGGTAT
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                              Left Arm
   901  TGAAAAATAT AGTAATGTAC ATATTTCTAA TGTTAACATA TTTATAGGTA
        ACTTTTTATA TCATTACATG TATAAAGATT ACAATTGTAT AAATATCCAT
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                              Left Arm
   951  AATCCAGGAA GGGTAATTTT TACATATCTA TATACGCTTA TTACAGTTAT
        TTAGGTCCTT CCCATTAAAA ATGTATAGAT ATATGCGAAT AATGTCAATA
```

```
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1001      TAAAAATATA CTTGCAAACA TGTTAGAAGT AAAAAAGAAA GAACTAATTT
                ATTTTTATAT GAACGTTTGT ACAATCTTCA TTTTTTCTTT CTTGATTAAA
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1051      TACAAAGTGC TTTACCAAAA TGCCAATGGA AATTACTTAG TATGTATATA
                ATGTTTCACG AAATGGTTTT ACGGTTACCT TTAATGAATC ATACATATAT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1101      ATGTATAAAG GTATGAATAT CACAAACAGC AAATCGGCTA TTCCCAAGTT
                TACATATTTC CATACTTATA GTGTTTGTCG TTTAGCCGAT AAGGGTTCAA
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm

      1151      GAGAAACGGT ATAATAGATA TATTTCTAGA TACCATTAAT AACCTTATAA
                CTCTTTGCCA TATTATCTAT ATAAAGATCT ATGGTAATTA TTGGAATATT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1201      GCTTGACGTT TCCTATAATG CCTACTAAGA AAACTAGAAG ATACATACAT
                CGAACTGCAA AGGATATTAC GGATGATTCT TTTGATCTTC TATGTATGTA
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1251      ACTAACGCCA TACGAGAGTA ACTACTCATC GTATAACTAC TGTTGCTAAC
                TGATTGCGGT ATGCTCTCAT TGATGAGTAG CATATTGATG ACAACGATTG
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1301      AGTGACACTG ATGTTATAAC TCATCTTTGA TGTGGTATAA ATGTATAATA
                TCACTGTGAC TACAATATTG AGTAGAAACT ACACCATATT TACATATTAT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1351      ACTATATTAC ACTGGTATTT TATTTCAGTT ATATACTATA TAGTATTAAA
                TGATATAATG TGACCATAAA ATAAAGTCAA TATATGATAT ATCATAATTT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1401      AATTATATTT GTATAATTAT ATTATTATAT TCAGTGTAGA AAGTAAAATA
                TTAATATAAA CATATTAATA TAATAATATA AGTCACATCT TTCATTTTAT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1451      CTATAAATAT GTATCTCTTA TTTATAACTT ATTAGTAAAG TATGTACTAT
                GATATTTATA CATAGAGAAT AAATATTGAA TAATCATTTC ATACATGATA
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1501      TCAGTTATAT TGTTTTATAA AAGCTAAATG CTACTAGATT GATATAAATG
                AGTCAATATA ACAAAATATT TTCGATTTAC GATGATCTAA CTATATTTAC
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
      1551      AATATGTAAT AAATTAGTAA TGTAGTATAC TAATATTAAC TCACATTTGA
                TTATACATTA TTTAATCATT ACATCATATG ATTATAATTG AGTGTAAACT
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Left Arm
                                                                30K Pr.
                                                                ~~~~
      1601      CTAATTAGCT ATAAAAACCC TAAGGTAGGC GGCCGCACTA GAGGATTCGA
                GATTAATCGA TATTTTTGGG ATTCCATCCG CCGGCGTGAT CTCCTAAGCT
```

```
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1651    CAAACACCAA TAATTCCCTT CTCTTCATTC CGGACATTAA ATTGGCTATA
                GTTTGTGGTT ATTAAGGGAA GAGAAGTAAG GCCTGTAATT TAACCGATAT
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1701    GATAATAAAG ACATTGAGAT GTTACAGGCT CTGTTCAAAT ACGACATTAA
                CTATTATTTC TGTAACTCTA CAATGTCCGA GACAAGTTTA TGCTGTAATT
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1751    TATCTATTCT GCTAATCTGG AAAATGTACT ATTGGATGAT GCCGAAATAG
                ATAGATAAGA CGATTAGACC TTTTACATGA TAACCTACTA CGGCTTTATC
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1801    CTAAAATGAT TATAGAAAAG CATGTTGAAT ACAAGTCTGA CTCCTATACA
                GATTTTACTA ATATCTTTTC GTACAACTTA TGTTCAGACT GAGGATATGT
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1851    AAAGATCTCG ATATAGTCAA GAATAATAAA TTGGATGAAA TAATTAGCAA
                TTTCTAGAGC TATATCAGTT CTTATTATTT AACCTACTTT ATTAATCGTT
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1901    AAACAAGGAA CTCAGACTCA TGTACGTCAA TTGTGTAAAG AAAAACTAAT
                TTTGTTCCTT GAGTCTGAGT ACATGCAGTT AACACATTTC TTTTTGATTA
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        1951    TAGATTCTCC CACATTTTTG TTAACATTAC ACTAACTAAT TGGTAAAATT
                ATCTAAGAGG GTGTAAAAAC AATTGTAATG TGATTGATTA ACCATTTTAA
                          30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2001    GATAGAATAA TTATGTGTAT ATAAGATAGA TTTCCTATTG TCTTACTCAT
                CTATCTTATT AATACACATA TATTCTATCT AAAGGATAAC AGAATGAGTA
                     30K Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                                        hLFA-3
                                                      ~~~~~~~~~~~~~~~~~~
        2051    TGCATCGTGG GAATTCAGAT CAGCTTCCGC GGCATGGTTG CTGGGAGCGA
                ACGTAGCACC CTTAAGTCTA GTCGAAGGCG CCGTACCAAC GACCCTCGCT
```

```
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2101    CGCGGGGCGG GCCCTGGGGG TCCTCAGCGT GGTCTGCCTG CTGCACTGCT
                GCGCCCCGCC CGGGACCCCC AGGAGTCGCA CCAGACGGAC GACGTGACGA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2151    TTGGTTTCAT CAGCTGTTTT TCCCAACAAA TATATGGTGT TGTGTATGGG
                AACCAAAGTA GTCGACAAAA AGGGTTGTTT ATATACCACA ACACATACCC
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2201    AATGTAACTT TCCATGTACC AAGCAATGTG CCTTTAAAAG AGGTCCTATG
                TTACATTGAA AGGTACATGG TTCGTTACAC GGAAATTTTC TCCAGGATAC
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2251    GAAAAAACAA AAGGATAAAG TTGCAGAACT GGAAAATTCT GAATTCAGAG
                CTTTTTTGTT TTCCTATTTC AACGTCTTGA CCTTTTAAGA CTTAAGTCTC
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2301    CTTTCTCATC TTTTAAAAAT AGGGTTTATT TAGACACTGT GTCAGGTAGC
                GAAAGAGTAG AAAATTTTTA TCCCAAATAA ATCTGTGACA CAGTCCATCG
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2351    CTCACTATCT ACAACTTAAC ATCATCAGAT GAAGATGAGT ATGAAATGGA
                GAGTGATAGA TGTTGAATTG TAGTAGTCTA CTTCTACTCA TACTTTACCT
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2401    ATCGCCAAAT ATTACTGATA CCATGAAGTT CTTTCTTTAT GTGCTTGAGT
                TAGCGGTTTA TAATGACTAT GGTACTTCAA GAAAGAAATA CACGAACTCA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2451    CTCTTCCATC TCCCACACTA ACTTGTGCAT TGACTAATGG AAGCATTGAA
                GAGAAGGTAG AGGGTGTGAT TGAACACGTA ACTGATTACC TTCGTAACTT
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2501    GTCCAATGCA TGATACCAGA GCATTACAAC AGCCATCGAG GACTTATAAT
                CAGGTTACGT ACTATGGTCT CGTAATGTTG TCGGTAGCTC CTGAATATTA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2551    GTACTCATGG GATTGTCCTA TGGAGCAATG TAAACGTAAC TCAACCAGTA
                CATGAGTACC CTAACAGGAT ACCTCGTTAC ATTTGCATTG AGTTGGTCAT
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2601    TATATTTTAA GATGGAAAAT GATCTTCCAC AAAAAATACA GTGTACTCTT
                ATATAAAATT CTACCTTTTA CTAGAAGGTG TTTTTTATGT CACATGAGAA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2651    AGCAATCCAT TATTTAATAC AACATCATCA ATCATTTTGA CAACCTGTAT
                TCGTTAGGTA ATAAATTATG TTGTAGTAGT TAGTAAAACT GTTGGACATA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2701    CCCAAGCAGC GGTCATTCAA GACACAGATA TGCACTTATA CCCATACCAT
                GGGTTCGTCG CCAGTAAGTT CTGTGTCTAT ACGTGAATAT GGGTATGGTA
                                hLFA-3
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2751    TAGCAGTAAT TACAACATGT ATTGTGCTGT ATATGAATGG TATTCTGAAA
                ATCGTCATTA ATGTTGTACA TAACACGACA TATACTTACC ATAAGACTTT
```

```
                    hLFA-3                                  I3 Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~    ~~~~~~~~~~
        2801    TGTGACAGAA AACCAGACAG AACCAACTCC AATTGATTGG CTCGACCGGG
                ACACTGTCTT TTGGTCTGTC TTGGTTGAGG TTAACTAACC GAGCTGGCCC
                                I3 Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2851    AATGTACTAT CTACGTACGA AACCCGCATC CGCTCCCATT CAATTCACAT
                TTACATGATA GATGCATGCT TTGGGCGTAG GCGAGGGTAA GTTAAGTGTA
                                I3 Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2901    TGGACAAGGA TAAAATAAAA CCACTGGTGG TTTGCGATTC CGAAATCTGT
                ACCTGTTCCT ATTTTATTTT GGTGACCACC AAACGCTAAG GCTTTAGACA
                                I3 Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        2951    ACATCATGCA GTGGTTAAAC AAAAACATTT TTATTCTCAA ATGAGATAAA
                TGTAGTACGT CACCAATTTG TTTTTGTAAA AATAAGAGTT TACTCTATTT
                                I3 Pr.
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3001    GTGAAAATAT ATATCATTAT ATTACAAAGT ACAATTATTT AGGTTTAATC
                CACTTTTATA TATAGTAATA TAATGTTTCA TGTTAATAAA TCCAAATTAG
                  I3 Pr.                    hICAM
                ~~~~~~~~~~~   ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3051    AATCCCGCGG GCTATGGCTC CCAGCAGCCC CCGGCCCGCG CTGCCCGCAC
                TTAGGGCGCC CGATACCGAG GGTCGTCGGG GGCCGGGCGC GACGGGCGTG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3101    TCCTGGTCCT GCTCGGGGCT CTGTTCCCAG GACCTGGCAA TGCCCAGACA
                AGGACCAGGA CGAGCCCCGA GACAAGGGTC CTGGACCGTT ACGGGTCTGT
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3151    TCTGTGTCCC CCTCAAAAGT CATCCTGCCC CGGGGAGGCT CCGTGCTGGT
                AGACACAGGG GGAGTTTTCA GTAGGACGGG GCCCCTCCGA GGCACGACCA
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3201    GACATGCAGC ACCTCCTGTG ACCAGCCCAA GTTGTTGGGC ATAGAGACCC
                CTGTACGTCG TGGAGGACAC TGGTCGGGTT CAACAACCCG TATCTCTGGG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3251    CGTTGCCTAA AAAGGAGTTG CTCCTGCCTG GGAACAACCG GAAGGTGTAT
                GCAACGGATT TTTCCTCAAC GAGGACGGAC CCTTGTTGGC CTTCCACATA
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3301    GAACTGAGCA ATGTGCAAGA AGATAGCCAA CCAATGTGCT ATTCAAACTG
                CTTGACTCGT TACACGTTCT TCTATCGGTT GGTTACACGA TAAGTTTGAC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3351    CCCTGATGGG CAGTCAACAG CTAAAACCTT CCTCACCGTG TACTGGACTC
                GGGACTACCC GTCAGTTGTC GATTTTGGAA GGAGTGGCAC ATGACCTGAG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3401    CAGAACGGGT GGAACTGGCA CCCCTCCCCT CTTGGCAGCC AGTGGGCAAG
                GTCTTGCCCA CCTTGACCGT GGGGAGGGGA GAACCGTCGG TCACCCGTTC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3451    AACCTTACCC TACGCTGCCA GGTGGAGGGT GGGGCACCCC GGGCCAACCT
                TTGGAATGGG ATGCGACGGT CCACCTCCCA CCCCGTGGGG CCCGGTTGGA
```

```
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3501    CACCGTGGTG CTGCTCCGTG GGGAGAAGGA GCTGAAACGG GAGCCAGCTG
                GTGGCACCAC GACGAGGCAC CCCTCTTCCT CGACTTTGCC CTCGGTCGAC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3551    TGGGGGAGCC CGCTGAGGTC ACGACCACGG TGCTGGTGAG GAGAGATCAC
                ACCCCCTCGG GCGACTCCAG TGCTGGTGCC ACGACCACTC CTCTCTAGTG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3601    CATGGAGCCA ATTTCTCGTG CCGCACTGAA CTGGACCTGC GGCCCCAAGG
                GTACCTCGGT TAAAGAGCAC GGCGTGACTT GACCTGGACG CCGGGGTTCC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3651    GCTGGAGCTG TTTGAGAACA CCTCGGCCCC CTACCAGCTC CAGACCTTTG
                CGACCTCGAC AAACTCTTGT GGAGCCGGGG GATGGTCGAG GTCTGGAAAC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3701    TCCTGCCAGC GACTCCCCCA CAACTTGTCA GCCCCCGGGT CCTAGAGGTG
                AGGACGGTCG CTGAGGGGGT GTTGAACAGT CGGGGGCCCA GGATCTCCAC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3751    GACACGCAGG GGACCGTGGT CTGTTCCCTG GACGGGCTGT TCCCAGTCTC
                CTGTGCGTCC CCTGGCACCA GACAAGGGAC CTGCCCGACA AGGGTCAGAG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3801    GGAGGCCCAG GTCCACCTGG CACTGGGGGA CCAGAGGTTG AACCCCACAG
                CCTCCGGGTC CAGGTGGACC GTGACCCCCT GGTCTCCAAC TTGGGGTGTC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3851    TCACCTATGG CAACGACTCC TTCTCGGCCA AGGCCTCAGT CAGTGTGACC
                AGTGGATACC GTTGCTGAGG AAGAGCCGGT TCCGGAGTCA GTCACACTGG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3901    GCAGAGGACG AGGGCACCCA GCGGCTGACG TGTGCAGTAA TACTGGGGAA
                CGTCTCCTGC TCCCGTGGGT CGCCGACTGC ACACGTCATT ATGACCCCTT
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3951    CCAGAGCCAG GAGACACTGC AGACAGTGAC CATCTACAGC TTTCCGGCGC
                GGTCTCGGTC CTCTGTGACG TCTGTCACTG GTAGATGTCG AAAGGCCGCG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4001    CCAACGTGAT TCTGACGAAG CCAGAGGTCT CAGAAGGGAC CGAGGTGACA
                GGTTGCACTA AGACTGCTTC GGTCTCCAGA GTCTTCCCTG GCTCCACTGT
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4051    GTGAAGTGTG AGGCCCACCC TAGAGCCAAG GTGACGCTGA ATGGGGTTCC
                CACTTCACAC TCCGGGTGGG ATCTCGGTTC CACTGCGACT TACCCCAAGG
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4101    AGCCCAGCCA CTGGGCCCGA GGGCCCAGCT CCTGCTGAAG GCCACCCCAG
                TCGGGTCGGT GACCCGGGCT CCCGGGTCGA GGACGACTTC CGGTGGGGTC
                                hICAM
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4151    AGGACAACGG GCGCAGCTTC TCCTGCTCTG CAACCCTGGA GGTGGCCGGC
                TCCTGTTGCC CGCGTCGAAG AGGACGAGAC GTTGGGACCT CCACCGGCCG
```

```
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4201  CAGCTTATAC ACAAGAACCA GACCCGGGAG CTTCGTGTCC TGTATGGCCC
          GTCGAATATG TGTTCTTGGT CTGGGCCCTC GAAGCACAGG ACATACCGGG
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4251  CCGACTGGAC GAGAGGGATT GTCCGGGAAA CTGGACGTGG CCAGAAAATT
          GGCTGACCTG CTCTCCCTAA CAGGCCCTTT GACCTGCACC GGTCTTTTAA
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4301  CCCAGCAGAC TCCAATGTGC CAGGCTTGGG GGAACCCATT GCCCGAGCTC
          GGGTCGTCTG AGGTTACACG GTCCGAACCC CCTTGGGTAA CGGGCTCGAG
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4351  AAGTGTCTAA AGGATGGCAC TTTCCCACTG CCCATCGGGG AATCAGTGAC
          TTCACAGATT TCCTACCGTG AAAGGGTGAC GGGTAGCCCC TTAGTCACTG
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4401  TGTCACTCGA GATCTTGAGG GCACCTACCT CTGTCGGGCC AGGAGCACTC
          ACAGTGAGCT CTAGAACTCC CGTGGATGGA GACAGCCCGG TCCTCGTGAG
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4451  AAGGGGAGGT CACCCGCGAG GTGACCGTGA ATGTGCTCTC CCCCCGGTAT
          TTCCCCTCCA GTGGGCGCTC CACTGGCACT TACACGAGAG GGGGGCCATA
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4501  GAGATTGTCA TCATCACTGT GGTAGCAGCC GCAGTCATAA TGGGCACTGC
          CTCTAACAGT AGTAGTGACA CCATCGTCGG CGTCAGTATT ACCCGTGACG
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4551  AGGCCTCAGC ACGTACCTCT ATAACCGCCA GCGGAAGATC AAGAAATACA
          TCCGGAGTCG TGCATGGAGA TATTGGCGGT CGCCTTCTAG TTCTTTATGT
                              hICAM
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4601  GACTACAACA GGCCCAAAAA GGGACCCCCA TGAAACCGAA CACACAAGCC
          CTGATGTTGT CCGGGTTTTT CCCTGGGGGT ACTTTGGCTT GTGTGTTCGG
             hICAM                                  sE/L Pr.
          ~~~~~~~~~~~~~                  ~~~~~~~~~~~~~~~~~~~~~~~~
    4651  ACGCCTCCCT GAGCATGCAT GTAGCTTAAA AATTGAAATT TTATTTTTTT
          TGCGGAGGGA CTCGTACGTA CATCGAATTT TTAACTTTAA AATAAAAAAA
             sE/L Pr.
          ~~~~~~~~~~~~~~~~~~
    4701  TTTTTGGAAT ATAAATAAGC TCGAAGTCGA AATTCCTGCA GCCCGGGGCC
          AAAAACCTTA TATTTATTCG AGCTTCAGCT TTAAGGACGT CGGGCCCCGG
                              hB7.1
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4751  ATGGGCCACA CACGGAGGCA GGGAACATCA CCATCCAAGT GTCCATACCT
          TACCCGGTGT GTGCCTCCGT CCCTTGTAGT GGTAGGTTCA CAGGTATGGA
                              hB7.1
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4801  CAATTTCTTT CAGCTCTTGG TGCTGGCTGG TCTTTCTCAC TTCTGTTCAG
          GTTAAAGAAA GTCGAGAACC ACGACCGACC AGAAAGAGTG AAGACAAGTC
                              hB7.1
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    4851  GTGTTATCCA CGTGACCAAG GAAGTGAAAG AAGTGGCAAC GCTGTCCTGT
          CACAATAGGT GCACTGGTTC CTTCACTTTC TTCACCGTTG CGACAGGACA
```

```
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
4901                GGTCACAATG TTTCTGTTGA AGAGCTGGCA CAAACTCGCA TCTACTGGCA
                    CCAGTGTTAC AAAGACAACT TCTCGACCGT GTTTGAGCGT AGATGACCGT
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
4951                AAAGGAGAAG AAAATGGTGC TGACTATGAT GTCTGGAGAC ATGAATATAT
                    TTTCCTCTTC TTTTACCACG ACTGATACTA CAGACCTCTG TACTTATATA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5001                GGCCCGAGTA CAAGAACCGG ACCATCTTTG ATATCACTAA TAACCTCTCC
                    CCGGGCTCAT GTTCTTGGCC TGGTAGAAAC TATAGTGATT ATTGGAGAGG
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5051                ATTGTGATCC TGGCTCTGCG CCCATCTGAC GAGGGCACAT ACGAGTGTGT
                    TAACACTAGG ACCGAGACGC GGGTAGACTG CTCCCGTGTA TGCTCACACA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5101                TGTTCTGAAG TATGAAAAAG ACGCTTTCAA GCGGGAACAC CTGGCTGAAG
                    ACAAGACTTC ATACTTTTTC TGCGAAAGTT CGCCCTTGTG GACCGACTTC
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5151                TGACGTTATC AGTCAAAGCT GACTTCCCTA CACCTAGTAT ATCTGACTTT
                    ACTGCAATAG TCAGTTTCGA CTGAAGGGAT GTGGATCATA TAGACTGAAA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5201                GAAATTCCAA CTTCTAATAT TAGAAGGATA ATTTGCTCAA CCTCTGGAGG
                    CTTTAAGGTT GAAGATTATA ATCTTCCTAT TAAACGAGTT GGAGACCTCC
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5251                TTTTCCAGAG CCTCACCTCT CCTGGTTGGA AAATGGAGAA GAATTAAATG
                    AAAAGGTCTC GGAGTGGAGA GGACCAACCT TTTACCTCTT CTTAATTTAC
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5301                CCATCAACAC AACAGTTTCC CAAGATCCTG AAACTGAGCT CTATGCTGTT
                    GGTAGTTGTG TTGTCAAAGG GTTCTAGGAC TTTGACTCGA GATACGACAA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5351                AGCAGCAAAC TGGATTTCAA TATGACAACC AACCACAGCT TCATGTGTCT
                    TCGTCGTTTG ACCTAAAGTT ATACTGTTGG TTGGTGTCGA AGTACACAGA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5401                CATCAAGTAT GGACATTTAA GAGTGAATCA GACCTTCAAC TGGAATACAA
                    GTAGTTCATA CCTGTAAATT CTCACTTAGT CTGGAAGTTG ACCTTATGTT
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5451                CCAAGCAAGA GCATTTTCCT GATAACCTGC TCCCATCCTG GGCCATTACC
                    GGTTCGTTCT CGTAAAAGGA CTATTGGACG AGGGTAGGAC CCGGTAATGG
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5501                TTAATCTCAG TAAATGGAAT TTTCGTGATA TGCTGCCTGA CCTACTGCTT
                    AATTAGAGTC ATTTACCTTA AAAGCACTAT ACGACGGACT GGATGACGAA
                              hB7.1
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5551                TGCCCCACGC TGCAGAGAGA GAAGGAGGAA TGAGAGATTG AGAAGGGAAA
                    ACGGGGTGCG ACGTCTCTCT CTTCCTCCTT ACTCTCTAAC TCTTCCCTTT
```

```
          hB7.1
          ~~~~~~~~~~~~~~~~~~
5601      GTGTACGCCC TGTATAAAAG CTTTCTAGGT TTTTGTTTAG GGCTGCAGGA
          CACATGCGGG ACATATTTTC GAAAGATCCA AAAACAAATC CCGACGTCCT
5651      ATTCCTCGAG GGATCCCGAT TTTTATGACT AGTTAATCAA ATAAAAAGCA
          TAAGGAGCTC CCTAGGGCTA AAAATACTGA TCAATTAGTT TATTTTTCGT
                                                          Right Arm
5701      TACAAGCTAT TGCTTCGCTA TCGTTACAAA ATGGCAGGAA TTTTGTGTAA
          ATGTTCGATA ACGAAGCGAT AGCAATGTTT TACCGTCCTT AAAACACATT
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
5751      ACTAAGCCAC ATACTTGCCA ATGAAAAAAA TAGTAGAAAG GATACTATTT
          TGATTCGGTG TATGAACGGT TACTTTTTTT ATCATCTTTC CTATGATAAA
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
5801      TAATGGGATT AGATGTTAAG GTTCCTTGGG ATTATAGTAA CTGGGCATCT
          ATTACCCTAA TCTACAATTC CAAGGAACCC TAATATCATT GACCCGTAGA
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
5851      GTTAACTTTT ACGACGTTAG GTTAGATACT GATGTTACAG ATTATAATAA
          CAATTGAAAA TGCTGCAATC CAATCTATGA CTACAATGTC TAATATTATT
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
5901      TGTTACAATA AAATACATGA CAGGATGTGA TATTTTTCCT CATATAACTC
          ACAATGTTAT TTTATGTACT GTCCTACACT ATAAAAAGGA GTATATTGAG
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
5951      TTGGAATAGC AAATATGGAT CAATGTGATA GATTTGAAAA TTTCAAAAAG
          AACCTTATCG TTTATACCTA GTTACACTAT CTAAACTTTT AAAGTTTTTC
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6001      CAAATAACTG ATCAAGATTT ACAGACTATT TCTATAGTCT GTAAAGAAGA
          GTTTATTGAC TAGTTCTAAA TGTCTGATAA AGATATCAGA CATTTCTTCT
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6051      GATGTGTTTT CCTCAGAGTA ACGCCTCTAA ACAGTTGGGA GCGAAAGGAT
          CTACACAAAA GGAGTCTCAT TGCGGAGATT TGTCAACCCT CGCTTTCCTA
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6101      GCGCTGTAGT TATGAAACTG GAGGTATCTG ATGAACTTAG AGCCCTAAGA
          CGCGACATCA ATACTTTGAC CTCCATAGAC TACTTGAATC TCGGGATTCT
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6151      AATGTTCTGC TGAATGCGGT ACCCTGTTCG AAGGACGTGT TTGGTGATAT
          TTACAAGACG ACTTACGCCA TGGGACAAGC TTCCTGCACA AACCACTATA
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6201      CACAGTAGAT AATCCGTGGA ATCCTCACAT AACAGTAGGA TATGTTAAGG
          GTGTCATCTA TTAGGCACCT TAGGAGTGTA TTGTCATCCT ATACAATTCC
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6251      AGGACGATGT CGAAAACAAG AAACGCCTAA TGGAGTGCAT GTCCAAGTTT
          TCCTGCTACA GCTTTTGTTC TTTGCGGATT ACCTCACGTA CAGGTTCAAA
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
```

```
6301  AGGGGGCAAG AAATACAAGT TCTAGGATGG TATTAATAAG TATCTAAGTA
      TCCCCCGTTC TTTATGTTCA AGATCCTACC ATAATTATTC ATAGATTCAT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6351  TTTGGTATAA TTTATTAAAT AGTATAATTA TAACAAATAA TAAATAACAT
      AAACCATATT AAATAATTTA TCATATTAAT ATTGTTTATT ATTTATTGTA
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6401  GATAACGGTT TTTATTAGAA TAAAATAGAG ATAATATCAT AATGATATAT
      CTATTGCCAA AAATAATCTT ATTTTATCTC TATTATAGTA TTACTATATA
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6451  AATACTTCAT TACCAGAAAT GAGTAATGGA AGACTTATAA ATGAACTGCA
      TTATGAAGTA ATGGTCTTTA CTCATTACCT TCTGAATATT TACTTGACGT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6501  TAAAGCTATA AGGTATAGAG ATATAAATTT AGTAAGGTAT ATACTTAAAA
      ATTTCGATAT TCCATATCTC TATATTTAAA TCATTCCATA TATGAATTTT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6551  AATGCAAATA CAATAACGTA AATATACTAT CAACGTCTTT GTATTTAGCC
      TTACGTTTAT GTTATTGCAT TTATATGATA GTTGCAGAAA CATAAATCGG
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6601  GTAAGTATTT CTGATATAGA AATGGTAAAA TTATTACTAG AACACGGTGC
      CATTCATAAA GACTATATCT TTACCATTTT AATAATGATC TTGTGCCACG
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6651  CGATATTTTA AAATGTAAAA ATCCTCCTCT TCATAAAGCT GCTAGTTTAG
      GCTATAAAAT TTTACATTTT TAGGAGGAGA AGTATTTCGA CGATCAAATC
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6701  ATAATACAGA AATTGCTAAA CTACTAATAG ATTCTGGCGC TGACATAGAA
      TATTATGTCT TTAACGATTT GATGATTATC TAAGACCGCG ACTGTATCTT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6751  CAGATACATT CTGGAAATAG TCCGTTATAT ATTTCTGTAT ATAGAAACAA
      GTCTATGTAA GACCTTTATC AGGCAATATA TAAAGACATA TATCTTTGTT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6801  TAAGTCATTA ACTAGATATT TATTAAAAAA AGGTGTTAAT TGTAATAGAT
      ATTCAGTAAT TGATCTATAA ATAATTTTTT TCCACAATTA ACATTATCTA
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6851  TCTTTCTAAA TTATTACGAT GTACTGTATG ATAAGATATC TGATGATATG
      AGAAAGATTT AATAATGCTA CATGACATAC TATTCTATAG ACTACTATAC
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6901  TATAAAATAT TTATAGATTT TAATATTGAT CTTAATATAC AAACTAGAAA
      ATATTTTATA AATATCTAAA ATTATAACTA GAATTATATG TTTGATCTTT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
6951  TTTTGAAACT CCGTTACATT ACGCTATAAA GTATAAGAAT ATAGATTTAA
      AAAACTTTGA GGCAATGTAA TGCGATATTT CATATTCTTA TATCTAAATT
      ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
```

```
7001   TTAGGATATT GTTAGATAAT AGTATTAAAA TAGATAAAAG TTTATTTTTG
       AATCCTATAA CAATCTATTA TCATAATTTT ATCTATTTTC AAATAAAAAC
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7051   CATAAACAGT ATCTCATAAA GGCACTTAAA AATAATTGTA GTTACGATAT
       GTATTTGTCA TAGAGTATTT CCGTGAATTT TTATTAACAT CAATGCTATA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7101   AATAGCGTTA CTTATAAATC ACGGAGTGCC TATAAACGAA CAAGATGATT
       TTATCGCAAT GAATATTTAG TGCCTCACGG ATATTTGCTT GTTCTACTAA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7151   TAGGTAAAAC CCCATTACAT CATTCGGTAA TTAATAGAAG AAAAGATGTA
       ATCCATTTTG GGGTAATGTA GTAAGCCATT AATTATCTTC TTTTCTACAT
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7201   ACAGCACTTC TGTTAAATCT AGGAGCTGAT ATAAACGTAA TAGATGACTG
       TGTCGTGAAG ACAATTTAGA TCCTCGACTA TATTTGCATT ATCTACTGAC
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7251   TATGGGCAGT CCCTTACATT ACGCTGTTTC ACGTAACGAT ATCGAAACAA
       ATACCCGTCA GGGAATGTAA TGCGACAAAG TGCATTGCTA TAGCTTTGTT
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7301   CAAAGACACT TTTAGAAAGA GGATCTAATG TTAATGTGGT TAATAATCAT
       GTTTCTGTGA AAATCTTTCT CCTAGATTAC AATTACACCA ATTATTAGTA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7351   ATAGATACCG TTCTAAATAT AGCTGTTGCA TCTAAAAACA AAACTATAGT
       TATCTATGGC AAGATTTATA TCGACAACGT AGATTTTTGT TTTGATATCA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7401   AAACTTATTA CTGAAGTACG GTACTGATAC AAAGTTGGTA GGATTAGATA
       TTTGAATAAT GACTTCATGC CATGACTATG TTTCAACCAT CCTAATCTAT
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7451   AACATGTTAT TCACATAGCT ATAGAAATGA AAGATATTAA TATACTGAAT
       TTGTACAATA AGTGTATCGA TATCTTTACT TTCTATAATT ATATGACTTA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7501   GCGATCTTAT TATATGGTTG CTATGTAAAC GTCTATAATC ATAAAGGTTT
       CGCTAGAATA ATATACCAAC GATACATTTG CAGATATTAG TATTTCCAAA
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7551   CACTCCTCTA TACATGGCAG TTAGTTCTAT GAAAACAGAA TTTGTTAAAC
       GTGAGGAGAT ATGTACCGTC AATCAAGATA CTTTTGTCTT AAACAATTTG
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7601   TCTTACTTGA CCACGGTGCT TACGTAAATG CTAAAGCTAA GTTATCTGGA
       AGAATGAACT GGTGCCACGA ATGCATTTAC GATTTCGATT CAATAGACCT
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                          Right Arm
7651   AATACTCCTT TACATAAAGC TATGTTATCT AATAGTTTTA ATAATATAAA
       TTATGAGGAA ATGTATTTCG ATACAATAGA TTATCAAAAT TATTATATTT
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
```

```
                                Right Arm
      7701   ATTACTTTTA TCTTATAACG CCGACTATAA TTCTCTAAAT AATCACGGTA
             TAATGAAAAT AGAATATTGC GGCTGATATT AAGAGATTTA TTAGTGCCAT
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      7751   ATACGCCTCT AACTTGTGTT AGCTTTTTAG ATGACAAGAT AGCTATTATG
             TATGCGGAGA TTGAACACAA TCGAAAAATC TACTGTTCTA TCGATAATAC
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      7801   ATAATATCTA AAATGATGTT AGAAATATCT AAAAATCCTG AAATAGCTAA
             TATTATAGAT TTTACTACAA TCTTTATAGA TTTTTAGGAC TTTATCGATT
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      7851   TTCAGAAGGT TTTATAGTAA ACATGGAACA TATAAACAGT AATAAAAGAC
             AAGTCTTCCA AAATATCATT TGTACCTTGT ATATTTGTCA TTATTTTCTG
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      7901   TACTATCTAT AAAAGAATCA TGCGAAAAAG AACTAGATGT TATAACACAT
             ATGATAGATA TTTTCTTAGT ACGCTTTTTC TTGATCTACA ATATTGTGTA
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      7951   ATAAAGTTAA ATTCTATATA TTCTTTTAAT ATCTTTCTTG ACAATAACAT
             TATTTCAATT TAAGATATAT AAGAAAATTA TAGAAAGAAC TGTTATTGTA
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8001   AGATCTTATG GTAAAGTTCG TAACTAATCC TAGAGTTAAT AAGATACCTG
             TCTAGAATAC CATTTCAAGC ATTGATTAGG ATCTCAATTA TTCTATGGAC
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8051   CATGTATACG TATATATAGG GAATTAATAC GGAAAAATAA ATCATTAGCT
             GTACATATGC ATATATATCC CTTAATTATG CCTTTTTATT TAGTAATCGA
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8101   TTTCATAGAC ATCAGCTAAT AGTTAAAGCT GTAAAAGAGA GTAAGAATCT
             AAAGTATCTG TAGTCGATTA TCAATTTCGA CATTTTCTCT CATTCTTAGA
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8151   AGGAATAATA GGTAGGTTAC CTATAGATAT CAAACATATA ATAATGGAAC
             TCCTTATTAT CCATCCAATG GATATCTATA GTTTGTATAT TATTACCTTG
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8201   TATTAAGTAA TAATGATTTA CATTCTGTTA TCACCAGCTG TTGTAACCCA
             ATAATTCATT ATTACTAAAT GTAAGACAAT AGTGGTCGAC AACATTGGGT
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
                                Right Arm
      8251   GTAGTATAAA GAGCTCCAGC TTTTGTTCCC TTTAGTGAGG GTTAATTCCG
             CATCATATTT CTCGAGGTCG AAAACAAGGG AAATCACTCC CAATTAAGGC
             ~~~~~~~~~~~~~~~~~
                Right Arm
      8301   AGCTTGGCGT AATCATGGTC ATAGCTGTTT CCTGTGTGAA ATTGTTATCC
             TCGAACCGCA TTAGTACCAG TATCGACAAA GGACACACTT TAACAATAGG
      8351   GCTCACAATT CCACACAACA TACGAGCCGG AAGCATAAAG TGTAAAGCCT
             CGAGTGTTAA GGTGTGTTGT ATGCTCGGCC TTCGTATTTC ACATTTCGGA
      8401   GGGGTGCCTA ATGAGTGAGC TAACTCACAT TAATTGCGTT GCGCTCACTG
             CCCCACGGAT TACTCACTCG ATTGAGTGTA ATTAACGCAA CGCGAGTGAC
      8451   CCCGCTTTCC AGTCGGGAAA CCTGTCGTGC CAGCTGCATT AATGAATCGG
```

```
      GGGCGAAAGG TCAGCCCTTT GGACAGCACG GTCGACGTAA TTACTTAGCC
8501  CCAACGCGCG GGGAGAGGCG GTTTGCGTAT TGGGCGCTCT TCCGCTTCCT
      GGTTGCGCGC CCCTCTCCGC CAAACGCATA ACCCGCGAGA AGGCGAAGGA
8551  CGCTCACTGA CTCGCTGCGC TCGGTCGTTC GGCTGCGGCG AGCGGTATCA
      GCGAGTGACT GAGCGACGCG AGCCAGCAAG CCGACGCCGC TCGCCATAGT
8601  GCTCACTCAA AGGCGGTAAT ACGGTTATCC ACAGAATCAG GGGATAACGC
      CGAGTGAGTT TCCGCCATTA TGCCAATAGG TGTCTTAGTC CCCTATTGCG
8651  AGGAAAGAAC ATGTGAGCAA AAGGCCAGCA AAAGGCCAGG AACCGTAAAA
      TCCTTTCTTG TACACTCGTT TTCCGGTCGT TTTCCGGTCC TTGGCATTTT
8701  AGGCCGCGTT GCTGGCGTTT TTCCATAGGC TCCGCCCCCC TGACGAGCAT
      TCCGGCGCAA CGACCGCAAA AAGGTATCCG AGGCGGGGGG ACTGCTCGTA
8751  CACAAAAATC GACGCTCAAG TCAGAGGTGG CGAAACCCGA CAGGACTATA
      GTGTTTTTAG CTGCGAGTTC AGTCTCCACC GCTTTGGGCT GTCCTGATAT
8801  AAGATACCAG GCGTTTCCCC CTGGAAGCTC CCTCGTGCGC TCTCCTGTTC
      TTCTATGGTC CGCAAAGGGG GACCTTCGAG GGAGCACGCG AGAGGACAAG
8851  CGACCCTGCC GCTTACCGGA TACCTGTCCG CCTTTCTCCC TTCGGGAAGC
      GCTGGGACGG CGAATGGCCT ATGGACAGGC GGAAAGAGGG AAGCCCTTCG
8901  GTGGCGCTTT CTCATAGCTC ACGCTGTAGG TATCTCAGTT CGGTGTAGGT
      CACCGCGAAA GAGTATCGAG TGCGACATCC ATAGAGTCAA GCCACATCCA
8951  CGTTCGCTCC AAGCTGGGCT GTGTGCACGA ACCCCCCGTT CAGCCCGACC
      GCAAGCGAGG TTCGACCCGA CACACGTGCT TGGGGGGCAA GTCGGGCTGG
9001  GCTGCGCCTT ATCCGGTAAC TATCGTCTTG AGTCCAACCC GGTAAGACAC
      CGACGCGGAA TAGGCCATTG ATAGCAGAAC TCAGGTTGGG CCATTCTGTG
9051  GACTTATCGC CACTGGCAGC AGCCACTGGT AACAGGATTA GCAGAGCGAG
      CTGAATAGCG GTGACCGTCG TCGGTGACCA TTGTCCTAAT CGTCTCGCTC
9101  GTATGTAGGC GGTGCTACAG AGTTCTTGAA GTGGTGGCCT AACTACGGCT
      CATACATCCG CCACGATGTC TCAAGAACTT CACCACCGGA TTGATGCCGA
9151  ACACTAGAAG GACAGTATTT GGTATCTGCG CTCTGCTGAA GCCAGTTACC
      TGTGATCTTC CTGTCATAAA CCATAGACGC GAGACGACTT CGGTCAATGG
9201  TTCGGAAAAA GAGTTGGTAG CTCTTGATCC GGCAAACAAA CCACCGCTGG
      AAGCCTTTTT CTCAACCATC GAGAACTAGG CCGTTTGTTT GGTGGCGACC
9251  TAGCGGTGGT TTTTTTGTTT GCAAGCAGCA GATTACGCGC AGAAAAAAAG
      ATCGCCACCA AAAAAACAAA CGTTCGTCGT CTAATGCGCG TCTTTTTTTC
9301  GATCTCAAGA AGATCCTTTG ATCTTTTCTA CGGGGTCTGA CGCTCAGTGG
      CTAGAGTTCT TCTAGGAAAC TAGAAAAGAT GCCCCAGACT GCGAGTCACC
9351  AACGAAAACT CACGTTAAGG GATTTTGGTC ATGAGATTAT CAAAAAGGAT
      TTGCTTTTGA GTGCAATTCC CTAAAACCAG TACTCTAATA GTTTTTCCTA
9401  CTTCACCTAG ATCCTTTTAA ATTAAAAATG AAGTTTTAAA TCAATCTAAA
      GAAGTGGATC TAGGAAAATT TAATTTTTAC TTCAAAATTT AGTTAGATTT
9451  GTATATATGA GTAAACTTGG TCTGACAGTT ACCAATGCTT AATCAGTGAG
      CATATATACT CATTTGAACC AGACTGTCAA TGGTTACGAA TTAGTCACTC
9501  GCACCTATCT CAGCGATCTG TCTATTTCGT TCATCCATAG TTGCCTGACT
      CGTGGATAGA GTCGCTAGAC AGATAAAGCA AGTAGGTATC AACGGACTGA
9551  CCCCGTCGTG TAGATAACTA CGATACGGGA GGGCTTACCA TCTGGCCCCA
      GGGGCAGCAC ATCTATTGAT GCTATGCCCT CCCGAATGGT AGACCGGGGT
9601  GTGCTGCAAT GATACCGCGA GACCCACGCT CACCGGCTCC AGATTTATCA
      CACGACGTTA CTATGGCGCT CTGGGTGCGA GTGGCCGAGG TCTAAATAGT
9651  GCAATAAACC AGCCAGCCGG AAGGGCCGAG CGCAGAAGTG GTCCTGCAAC
      CGTTATTTGG TCGGTCGGCC TTCCCGGCTC GCGTCTTCAC CAGGACGTTG
9701  TTTATCCGCC TCCATCCAGT CTATTAATTG TTGCCGGGAA GCTAGAGTAA
      AAATAGGCGG AGGTAGGTCA GATAATTAAC AACGGCCCTT CGATCTCATT
9751  GTAGTTCGCC AGTTAATAGT TTGCGCAACG TTGTTGCCAT TGCTACAGGC
      CATCAAGCGG TCAATTATCA AACGCGTTGC AACAACGGTA ACGATGTCCG
9801  ATCGTGGTGT CACGCTCGTC GTTTGGTATG GCTTCATTCA GCTCCGGTTC
      TAGCACCACA GTGCGAGCAG CAAACCATAC CGAAGTAAGT CGAGGCCAAG
```

```
 9851   CCAACGATCA AGGCGAGTTA CATGATCCCC CATGTTGTGC AAAAAAGCGG
        GGTTGCTAGT TCCGCTCAAT GTACTAGGGG GTACAACACG TTTTTTCGCC
 9901   TTAGCTCCTT CGGTCCTCCG ATCGTTGTCA GAAGTAAGTT GGCCGCAGTG
        AATCGAGGAA GCCAGGAGGC TAGCAACAGT CTTCATTCAA CCGGCGTCAC
 9951   TTATCACTCA TGGTTATGGC AGCACTGCAT AATTCTCTTA CTGTCATGCC
        AATAGTGAGT ACCAATACCG TCGTGACGTA TTAAGAGAAT GACAGTACGG
10001   ATCCGTAAGA TGCTTTTCTG TGACTGGTGA GTACTCAACC AAGTCATTCT
        TAGGCATTCT ACGAAAAGAC ACTGACCACT CATGAGTTGG TTCAGTAAGA
10051   GAGAATAGTG TATGCGGCGA CCGAGTTGCT CTTGCCCGGC GTCAATACGG
        CTCTTATCAC ATACGCCGCT GGCTCAACGA GAACGGGCCG CAGTTATGCC
10101   GATAATACCG CGCCACATAG CAGAACTTTA AAAGTGCTCA TCATTGGAAA
        CTATTATGGC GCGGTGTATC GTCTTGAAAT TTTCACGAGT AGTAACCTTT
10151   ACGTTCTTCG GGGCGAAAAC TCTCAAGGAT CTTACCGCTG TTGAGATCCA
        TGCAAGAAGC CCCGCTTTTG AGAGTTCCTA GAATGGCGAC AACTCTAGGT
10201   GTTCGATGTA ACCCACTCGT GCACCCAACT GATCTTCAGC ATCTTTTACT
        CAAGCTACAT TGGGTGAGCA CGTGGGTTGA CTAGAAGTCG TAGAAAATGA
10251   TTCACCAGCG TTTCTGGGTG AGCAAAAACA GGAAGGCAAA ATGCCGCAAA
        AAGTGGTCGC AAAGACCCAC TCGTTTTTGT CCTTCCGTTT TACGGCGTTT
10301   AAAGGGAATA AGGGCGACAC GGAAATGTTG AATACTCATA CTCTTCCTTT
        TTTCCCTTAT TCCCGCTGTG CCTTTACAAC TTATGAGTAT GAGAAGGAAA
10351   TTCAATATTA TTGAAGCATT TATCAGGGTT ATTGTCTCAT GAGCGGATAC
        AAGTTATAAT AACTTCGTAA ATAGTCCCAA TAACAGAGTA CTCGCCTATG
10401   ATATTTGAAT GTATTTAGAA AAATAAACAA ATAGGGGTTC CGCGCACATT
        TATAAACTTA CATAAATCTT TTTATTTGTT TATCCCCAAG GCGCGTGTAA
10451   TCCCCGAAAA GTGCCACCTG AGGGGCTTTT CACGGTGGAC
```

FIGURE 3: Donor plasmid p1132

```
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1   TGAATGTTAA ATGTTATACT TTGGATGAAG CTATAAATAT GCATTGGAAA
        ACTTACAATT TACAATATGA AACCTACTTC GATATTTATA CGTAACCTTT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   51   AATAATCCAT TTAAAGAAAG GATTCAAATA CTACAAAACC TAAGCGATAA
        TTATTAGGTA AATTTCTTTC CTAAGTTTAT GATGTTTTGG ATTCGCTATT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  101   TATGTTAACT AAGCTTATTC TTAACGACGC TTTAAATATA CACAAATAAA
        ATACAATTGA TTCGAATAAG AATTGCTGCG AAATTTATAT GTGTTTATTT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  151   CATAATTTTT GTATAACCTA ACAAATAACT AAAACATAAA AATAATAAAA
        GTATTAAAAA CATATTGGAT TGTTTATTGA TTTTGTATTT TTATTATTTT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  201   GGAAATGTAA TATCGTAATT ATTTTACTCA GGAATGGGGT TAAATATTTA
        CCTTTACATT ATAGCATTAA TAAAATGAGT CCTTACCCCA ATTTATAAAT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  251   TATCACGTGT ATATCTATAC TGTTATCGTA TACTCTTTAC AATTACTATT
        ATAGTGCACA TATAGATATG ACAATAGCAT ATGAGAAATG TTAATGATAA
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  301   ACGAATATGC AAGAGATAAT AAGATTACGT ATTTAAGAGA ATCTTGTCAT
        TGCTTATACG TTCTCTATTA TTCTAATGCA TAAATTCTCT TAGAACAGTA
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  351   GATAATTGGG TACGACATAG TGATAAATGC TATTTCGCAT CGTTACATAA
        CTATTAACCC ATGCTGTATC ACTATTTACG ATAAAGCGTA GCAATGTATT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  401   AGTCAGTTGG AAAGATGGAT TTGACAGATG TAACTTAATA GGTGCAAAAA
        TCAGTCAACC TTTCTACCTA AACTGTCTAC ATTGAATTAT CCACGTTTTT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  451   TGTTAAATAA CAGCATTCTA TCGGAAGATA GGATACCAGT TATATTATAC
        ACAATTTATT GTCGTAAGAT AGCCTTCTAT CCTATGGTCA ATATAATATG
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  501   AAAAATCACT GGTTGGATAA AACAGATTCT GCAATATTCG TAAAAGATGA
        TTTTTAGTGA CCAACCTATT TTGTCTAAGA CGTTATAAGC ATTTTCTACT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  551   AGATTACTGC GAATTTGTAA ACTATGACAA TAAAAAGCCA TTTATCTCAA
        TCTAATGACG CTTAAACATT TGATACTGTT ATTTTTCGGT AAATAGAGTT
                        C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
  601   CGACATCGTG TAATTCTTCC ATGTTTTATG TATGTGTTTC AGATATTATG
        GCTGTAGCAC ATTAAGAAGG TACAAAATAC ATACACAAAG TCTATAATAC
```

```
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          651   AGATTACTAT AAACTTTTTG TATACTTATA TTCCGTAAAC TATATTAATC
          5     TCTAATGATA TTTGAAAAAC ATATGAATAT AAGGCATTTG ATATAATTAG
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          701   ATGAAGAAAA TGAAAAAGTA TAGAAGCTGT TCACGAGCGG TTGTTGAAAA
                TACTTCTTTT ACTTTTTCAT ATCTTCGACA AGTGCTCGCC AACAACTTTT
          10                C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          751   CAACAAAATT ATACATTCAA GATGGCTTAC ATATACGTCT GTGAGGCTAT
                GTTGTTTTAA TATGTAAGTT CTACCGAATG TATATGCAGA CACTCCGATA
                          C5 Right Arm
          15    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          801   CATGGATAAT GACAATGCAT CTCTAAATAG GTTTTTGGAC AATGGATTCG
                GTACCTATTA CTGTTACGTA GAGATTTATC CAAAAACCTG TTACCTAAGC
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          20    851   ACCCTAACAC GGAATATGGT ACTCTACAAT CTCCTCTTGA AATGGCTGTA
                TGGGATTGTG CCTTATACCA TGAGATGTTA GAGGAGAACT TTACCGACAT
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          901   ATGTTCAAGA ATACCGAGGC TATAAAAATC TTGATGAGGT ATGGAGCTAA
          25    TACAAGTTCT TATGGCTCCG ATATTTTTAG AACTACTCCA TACCTCGATT
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          951   ACCTGTAGTT ACTGAATGCA CAACTTCTTG TCTGCATGAT GCGGTGTTGA
                TGGACATCAA TGACTTACGT GTTGAAGAAC AGACGTACTA CGCCACAACT
          30                C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          1001  GAGACGACTA CAAAATAGTG AAAGATCTGT TGAAGAATAA CTATGTAAAC
                CTCTGCTGAT GTTTTATCAC TTTCTAGACA ACTTCTTATT GATACATTTG
                          C5 Right Arm
          35    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          1051  AATGTTCTTT ACAGCGGAGG CTTTACTCCT TTGTGTTTGG CAGCTTACCT
                TTACAAGAAA TGTCGCCTCC GAAATGAGGA AACACAAACC GTCGAATGGA
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          40    1101  TAACAAAGTT AATTTGGTTA AACTTCTATT GGCTCATTCG GCGGATGTAG
                ATTGTTTCAA TTAAACCAAT TTGAAGATAA CCGAGTAAGC CGCCTACATC
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          1151  ATATTTCAAA CACGGATCGG TTAACTCCTC TACATATAGC CGTATCAAAT
          45    TATAAAGTTT GTGCCTAGCC AATTGAGGAG ATGTATATCG GCATAGTTTA
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          1201  AAAAATTTAA CAATGGTTAA ACTTCTATTG AACAAAGGTG CTGATACTGA
                TTTTTAAATT GTTACCAATT TGAAGATAAC TTGTTTCCAC GACTATGACT
          50                C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          1251  CTTGCTGGAT AACATGGGAT GTACTCCTTT AATGATCGCT GTACAATCTG
                GAACGACCTA TTGTACCCTA CATGAGGAAA TTACTAGCGA CATGTTAGAC

          55
```

```
                         C5 Right Arm
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1301 GAAATATTGA AATATGTAGC ACACTACTTA AAAAAAATAA AATGTCCAGA
        CTTTATAACT TTATACATCG TGTGATGAAT TTTTTTTATT TTACAGGTCT
                         C5 Right Arm
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1351 ACTGGGAAAA ATTGATCTTG CCAGCTGTAA TTCATGGTAG AAAAGAAGTG
        TGACCCTTTT TAACTAGAAC GGTCGACATT AAGTACCATC TTTTCTTCAC
                         C5 Right Arm
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1401 CTCAGGCTAC TTTTCAACAA AGGAGCAGAT GTAAACTACA TCTTTGAAAG
        GAGTCCGATG AAAAGTTGTT TCCTCGTCTA CATTTGATGT AGAAACTTTC
                         C5 Right Arm
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1451 AAATGGAAAA TCATATACTG TTTTGGAATT GATTAAAGAA AGTTACTCTG
        TTTACCTTTT AGTATATGAC AAAACCTTAA CTAATTTCTT TCAATGAGAC
                  C5 Right Arm
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1501 AGACACAAAA GAGGTAGCTG AAGTGGTACT CTCAAAGGTA CGTGACTAAT
        TCTGTGTTTT CTCCATCGAC TTCACCATGA GAGTTTCCAT GCACTGATTA
                                                Repeat Region
                                                ~~~~~~~~~~
   1551 TAGCTATAAA AAGGATCGGC CGCTCTAGAA CTAGTGGATC GGGTTCTTTA
        ATCGATATTT TTCCTAGCCG GCGAGATCTT GATCACCTAG CCCAAGAAAT
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1601 TTCTATACTT AAAAAGTGAA AATAAATACA AAGGTTCTTG AGGGTTGTGT
        AAGATATGAA TTTTTCACTT TTATTTATGT TTCCAAGAAC TCCCAACACA
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1651 TAAATTGAAA GCGAGAAATA ATCATAAATT ATTTCATTAT CGCGATATCC
        ATTTAACTTT CGCTCTTTAT TAGTATTTAA TAAAGTAATA GCGCTATAGG
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1701 GTTAAGTTTG TATCGTACCC CGATCCCCCG AGCCATGCAG GCCGAAGGCC
        CAATTCAAAC ATAGCATGGG GCTAGGGGGC TCGGTACGTC CGGCTTCCGG
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1751 GGGGCACAGG GGGTTCGACG GGCGATGCTG ATGGCCCAGG AGGCCCTGGC
        CCCCGTGTCC CCCAAGCTGC CCGCTACGAC TACCGGGTCC TCCGGGACCG
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1801 ATTCCTGATG GCCCAGGGGG CAATGCTGGC GGCCCAGGAG AGGCGGGTGC
        TAAGGACTAC CGGGTCCCCC GTTACGACCG CCGGGTCCTC TCCGCCCACG
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1851 CACGGGCGGC AGAGGTCCCC GGGGCGCAGG GGCAGCAAGG GCCTCGGGGC
        GTGCCCGCCG TCTCCAGGGG CCCCGCGTCC CCGTCGTTCC CGGAGCCCCG
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1901 CGGGAGGAGG CGCCCCGCGG GGTCCGCATG GCGGCGCGGC TTCAGGGCTG
        GCCCTCCTCC GCGGGGCGCC CCAGGCGTAC CGCCGCGCCG AAGTCCCGAC
                        Repeat Region
        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
   1951 AATGGATGCT GCAGATGCGG GGCCAGGGGG CCGGAGAGCC GCCTGCTTGA
        TTACCTACGA CGTCTACGCC CCGGTCCCCC GGCCTCTCGG CGGACGAACT
```

```
                Repeat Region
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2001     GTTCTACCTC GCCATGCCTT TCGCGACACC CATAGCTTGA TATCGAATTC
               CAAGATGGAG CGGTACGGAA AGCGCTGTGG GTATCGAACT ATAGCTTAAG
                                                 C1B promoter
                                               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2051     TAGGGGGATC CACTAGTTCT AGAGGATCAT TATTTAACGT AAACTAAATG
               ATCCCCCTAG GTGATCAAGA TCTCCTAGTA ATAAATTGCA TTTGATTTAC
                              C1B promoter
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2101     GAAAAGCTAT TTACAGGTAC ATACGGTGTT TTTCTGGAAT CAAATGATTC
               CTTTTCGATA AATGTCCATG TATGCCACAA AAAGACCTTA GTTTACTAAG
                              C1B promoter
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2151     TGATTTTGAG GATTTTATCA ATACAATAAT GACAGTGCTA ACTGGTAAAA
               ACTAAAACTC CTAAAATAGT TATGTTATTA CTGTCACGAT TGACCATTTT
                              C1B promoter
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2201     AAGAAAGCAA ACAATTATCA TGGCTAACAA TTTTTATTAT ATTTGTAGTA
               TTCTTTCGTT TGTTAATAGT ACCGATTGTT AAAAATAATA TAAACATCAT
                              C1B promoter
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2251     TGCATAGTGG TCTTTACGTT TCTTTATTTA AAGTTAATGT GTTAAGATTA
               ACGTATCACC AGAAATGCAA AGAAATAAAT TTCAATTACA CAATTCTAAT
                C1B promoter                          LacZ
               ~~~~~~~~~~~~~~~~~~~          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2301     AATGGAGTAA TTGGATCCCC CATCGATGGG GAATTCACTG GCCGTCGTTT
               TTACCTCATT AACCTAGGGG GTAGCTACCC CTTAAGTGAC CGGCAGCAAA
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2351     TACAACGTCG TGACTGGGAA AACCCTGGCG TTACCCAACT TAATCGCCTT
               ATGTTGCAGC ACTGACCCTT TTGGGACCGC AATGGGTTGA ATTAGCGGAA
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2401     GCAGCACATC CCCCTTTCGC CAGCTGGCGT AATAGCGAAG AGGCCCGCAC
               CGTCGTGTAG GGGGAAAGCG GTCGACCGCA TTATCGCTTC TCCGGGCGTG
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2451     CGATCGCCCT TCCCAACAGT TGCGCAGCCT GAATGGCGAA TGGCGCTTTG
               GCTAGCGGGA AGGGTTGTCA ACGCGTCGGA CTTACCGCTT ACCGCGAAAC
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2501     CCTGGTTTCC GGCACCAGAA GCGGTGCCGG AAAGCTGGCT GGAGTGCGAT
               GGACCAAAGG CCGTGGTCTT CGCCACGGCC TTTCGACCGA CCTCACGCTA
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2551     CTTCCTGAGG CCGATACTGT CGTCGTCCCC TCAAACTGGC AGATGCACGG
               GAAGGACTCC GGCTATGACA GCAGCAGGGG AGTTTGACCG TCTACGTGCC
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2601     TTACGATGCG CCCATCTACA CCAACGTGAC CTATCCCATT ACGGTCAATC
               AATGCTACGC GGGTAGATGT GGTTGCACTG GATAGGGTAA TGCCAGTTAG
                                     LacZ
               ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2651     CGCCGTTTGT TCCCACGGAG AATCCGACGG GTTGTTACTC GCTCACATTT
               GCGGCAAACA AGGGTGCCTC TTAGGCTGCC CAACAATGAG CGAGTGTAAA
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2701      AATGTTGATG AAAGCTGGCT ACAGGAAGGC CAGACGCGAA TTATTTTTGA
                TTACAACTAC TTTCGACCGA TGTCCTTCCG GTCTGCGCTT AATAAAAACT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2751      TGGCGTTAAC TCGGCGTTTC ATCTGTGGTG CAACGGGCGC TGGGTCGGTT
                ACCGCAATTG AGCCGCAAAG TAGACACCAC GTTGCCCGCG ACCCAGCCAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2801      ACGGCCAGGA CAGTCGTTTG CCGTCTGAAT TTGACCTGAG CGCATTTTTA
                TGCCGGTCCT GTCAGCAAAC GGCAGACTTA AACTGGACTC GCGTAAAAAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2851      CGCGCCGGAG AAAACCGCCT CGCGGTGATG GTGCTGCGCT GGAGTGACGG
                GCGCGGCCTC TTTTGGCGGA GCGCCACTAC CACGACGCGA CCTCACTGCC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2901      CAGTTATCTG GAAGATCAGG ATATGTGGCG GATGAGCGGC ATTTTCCGTG
                GTCAATAGAC CTTCTAGTCC TATACACCGC CTACTCGCCG TAAAAGGCAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2951      ACGTCTCGTT GCTGCATAAA CCGACTACAC AAATCAGCGA TTTCCATGTT
                TGCAGAGCAA CGACGTATTT GGCTGATGTG TTTAGTCGCT AAAGGTACAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3001      GCCACTCGCT TTAATGATGA TTTCAGCCGC GCTGTACTGG AGGCTGAAGT
                CGGTGAGCGA AATTACTACT AAAGTCGGCG CGACATGACC TCCGACTTCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3051      TCAGATGTGC GGCGAGTTGC GTGACTACCT ACGGGTAACA GTTTCTTTAT
                AGTCTACACG CCGCTCAACG CACTGATGGA TGCCCATTGT CAAAGAAATA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3101      GGCAGGGTGA AACGCAGGTC GCCAGCGGCA CCGCGCCTTT CGGCGGTGAA
                CCGTCCCACT TTGCGTCCAG CGGTCGCCGT GGCGCGGAAA GCCGCCACTT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3151      ATTATCGATG AGCGTGGTGG TTATGCCGAT CGCGTCACAC TACGTCTGAA
                TAATAGCTAC TCGCACCACC AATACGGCTA GCGCAGTGTG ATGCAGACTT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3201      CGTCGAAAAC CCGAAACTGT GGAGCGCCGA AATCCCGAAT CTCTATCGTG
                GCAGCTTTTG GGCTTTGACA CCTCGCGGCT TTAGGGCTTA GAGATAGCAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3251      CGGTGGTTGA ACTGCACACC GCCGACGGCA CGCTGATTGA AGCAGAAGCC
                GCCACCAACT TGACGTGTGG CGGCTGCCGT GCGACTAACT TCGTCTTCGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3301      TGCGATGTCG GTTTCCGCGA GGTGCGGATT GAAAATGGTC TGCTGCTGCT
                ACGCTACAGC CAAAGGCGCT CCACGCCTAA CTTTTACCAG ACGACGACGA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3351      GAACGGCAAG CCGTTGCTGA TTCGAGGCGT TAACCGTCAC GAGCATCATC
                CTTGCCGTTC GGCAACGACT AAGCTCCGCA ATTGGCAGTG CTCGTAGTAG
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3401    CTCTGCATGG TCAGGTCATG GATGAGCAGA CGATGGTGCA GGATATCCTG
                GAGACGTACC AGTCCAGTAC CTACTCGTCT GCTACCACGT CCTATAGGAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3451    CTGATGAAGC AGAACAACTT TAACGCCGTG CGCTGTTCGC ATTATCCGAA
                GACTACTTCG TCTTGTTGAA ATTGCGGCAC GCGACAAGCG TAATAGGCTT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3501    CCATCCGCTG TGGTACACGC TGTGCGACCG CTACGGCCTG TATGTGGTGG
                GGTAGGCGAC ACCATGTGCG ACACGCTGGC GATGCCGGAC ATACACCACC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3551    ATGAAGCCAA TATTGAAACC CACGGCATGG TGCCAATGAA TCGTCTGACC
                TACTTCGGTT ATAACTTTGG GTGCCGTACC ACGGTTACTT AGCAGACTGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3601    GATGATCCGC GCTGGCTACC GGCGATGAGC GAACGCGTAA CGCGAATGGT
                CTACTAGGCG CGACCGATGG CCGCTACTCG CTTGCGCATT GCGCTTACCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3651    GCAGCGCGAT CGTAATCACC CGAGTGTGAT CATCTGGTCG CTGGGGAATG
                CGTCGCGCTA GCATTAGTGG GCTCACACTA GTAGACCAGC GACCCCTTAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3701    AATCAGGCCA CGGCGCTAAT CACGACGCGC TGTATCGCTG GATCAAATCT
                TTAGTCCGGT GCCGCGATTA GTGCTGCGCG ACATAGCGAC CTAGTTTAGA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3751    GTCGATCCTT CCCGCCCGGT GCAGTATGAA GGCGGCGGAG CCGACACCAC
                CAGCTAGGAA GGGCGGGCCA CGTCATACTT CCGCCGCCTC GGCTGTGGTG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3801    GGCCACCGAT ATTATTTGCC CGATGTACGC GCGCGTGGAT GAAGACCAGC
                CCGGTGGCTA TAATAAACGG GCTACATGCG CGCGCACCTA CTTCTGGTCG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3851    CCTTCCCGGC TGTGCCGAAA TGGTCCATCA AAAAATGGCT TTCGCTACCT
                GGAAGGGCCG ACACGGCTTT ACCAGGTAGT TTTTTACCGA AAGCGATGGA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3901    GGAGAGACGC GCCCGCTGAT CCTTTGCGAA TACGCCCACG CGATGGGTAA
                CCTCTCTGCG CGGGCGACTA GGAAACGCTT ATGCGGGTGC GCTACCCATT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        3951    CAGTCTTGGC GGTTTCGCTA AATACTGGCA GGCGTTTCGT CAGTATCCCC
                GTCAGAACCG CCAAAGCGAT TTATGACCGT CCGCAAAGCA GTCATAGGGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4001    GTTTACAGGG CGGCTTCGTC TGGGACTGGG TGGATCAGTC GCTGATTAAA
                CAAATGTCCC GCCGAAGCAG ACCCTGACCC ACCTAGTCAG CGACTAATTT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4051    TATGATGAAA ACGGCAACCC GTGGTCGGCT TACGGCGGTG ATTTTGGCGA
                ATACTACTTT TGCCGTTGGG CACCAGCCGA ATGCCGCCAC TAAAACCGCT
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4101      TACGCCGAAC GATCGCCAGT TCTGTATGAA CGGTCTGGTC TTTGCCGACC
                ATGCGGCTTG CTAGCGGTCA AGACATACTT GCCAGACCAG AAACGGCTGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4151      GCACGCCGCA TCCAGCGCTG ACGGAAGCAA AACACCAGCA GCAGTTTTTC
                CGTGCGGCGT AGGTCGCGAC TGCCTTCGTT TTGTGGTCGT CGTCAAAAAG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4201      CAGTTCCGTT TATCCGGGCA AACCATCGAA GTGACCAGCG AATACCTGTT
                GTCAAGGCAA ATAGGCCCGT TTGGTAGCTT CACTGGTCGC TTATGGACAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4251      CCGTCATAGC GATAACGAGC TCCTGCACTG GATGGTGGCG CTGGATGGTA
                GGCAGTATCG CTATTGCTCG AGGACGTGAC CTACCACCGC GACCTACCAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4301      AGCCGCTGGC AAGCGGTGAA GTGCCTCTGG ATGTCGCTCC ACAAGGTAAA
                TCGGCGACCG TTCGCCACTT CACGGAGACC TACAGCGAGG TGTTCCATTT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4351      CAGTTGATTG AACTGCCTGA ACTACCGCAG CCGGAGAGCG CCGGGCAACT
                GTCAACTAAC TTGACGGACT TGATGGCGTC GGCCTCTCGC GGCCCGTTGA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4401      CTGGCTCACA GTACGCGTAG TGCAACCGAA CGCGACCGCA TGGTCAGAAG
                GACCGAGTGT CATGCGCATC ACGTTGGCTT GCGCTGGCGT ACCAGTCTTC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4451      CCGGGCACAT CAGCGCCTGG CAGCAGTGGC GTCTGGCGGA AAACCTCAGT
                GGCCCGTGTA GTCGCGGACC GTCGTCACCG CAGACCGCCT TTTGGAGTCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4501      GTGACGCTCC CCGCCGCGTC CCACGCCATC CCGCATCTGA CCACCAGCGA
                CACTGCGAGG GGCGGCGCAG GGTGCGGTAG GGCGTAGACT GGTGGTCGCT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4551      AATGGATTTT TGCATCGAGC TGGGTAATAA GCGTTGGCAA TTTAACCGCC
                TTACCTAAAA ACGTAGCTCG ACCCATTATT CGCAACCGTT AAATTGGCGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4601      AGTCAGGCTT TCTTTCACAG ATGTGGATTG GCGATAAAAA ACAACTGCTG
                TCAGTCCGAA AGAAAGTGTC TACACCTAAC CGCTATTTTT TGTTGACGAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4651      ACGCCGCTGC GCGATCAGTT CACCCGTGCA CCGCTGGATA ACGACATTGG
                TGCGGCGACG CGCTAGTCAA GTGGGCACGT GGCGACCTAT TGCTGTAACC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4701      CGTAAGTGAA GCGACCCGCA TTGACCCTAA CGCCTGGGTC GAACGCTGGA
                GCATTCACTT CGCTGGGCGT AACTGGGATT GCGGACCCAG CTTGCGACCT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4751      AGGCGGCGGG CCATTACCAG GCCGAAGCAG CGTTGTTGCA GTGCACGGCA
                TCCGCCGCCC GGTAATGGTC CGGCTTCGTC GCAACAACGT CACGTGCCGT
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4801    GATACACTTG CTGATGCGGT GCTGATTACG ACCGCTCACG CGTGGCAGCA
                CTATGTGAAC GACTACGCCA CGACTAATGC TGGCGAGTGC GCACCGTCGT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4851    TCAGGGGAAA ACCTTATTTA TCAGCCGGAA AACCTACCGG ATTGATGGTA
                AGTCCCCTTT TGGAATAAAT AGTCGGCCTT TTGGATGGCC TAACTACCAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4901    GTGGTCAAAT GGCGATTACC GTTGATGTTG AAGTGGCGAG CGATACACCG
                CACCAGTTTA CCGCTAATGG CAACTACAAC TTCACCGCTC GCTATGTGGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4951    CATCCGGCGC GGATTGGCCT GAACTGCCAG CTGGCGCAGG TAGCAGAGCG
                GTAGGCCGCG CCTAACCGGA CTTGACGGTC GACCGCGTCC ATCGTCTCGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5001    GGTAAACTGG CTCGGATTAG GGCCGCAAGA AAACTATCCC GACCGCCTTA
                CCATTTGACC GAGCCTAATC CCGGCGTTCT TTTGATAGGG CTGGCGGAAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5051    CTGCCGCCTG TTTTGACCGC TGGGATCTGC CATTGTCAGA CATGTATACC
                GACGGCGGAC AAAACTGGCG ACCCTAGACG GTAACAGTCT GTACATATGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5101    CCGTACGTCT TCCCGAGCGA AAACGGTCTG CGCTGCGGGA CGCGCGAATT
                GGCATGCAGA AGGGCTCGCT TTTGCCAGAC GCGACGCCCT GCGCGCTTAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5151    GAATTATGGC CCACACCAGT GGCGCGGCGA CTTCCAGTTC AACATCAGCC
                CTTAATACCG GGTGTGGTCA CCGCGCCGCT GAAGGTCAAG TTGTAGTCGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5201    GGTACAGTCA ACAGCAATTG ATGGAAACCA GCCATTCGCC ATCTGCTGCA
                CCATGTCAGT TGTCGTTAAC TACCTTTGGT CGGTAAGCGG TAGACGACGT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5251    CGCGGAAGAG GCACATGGCT GAATATCGAC GGTTTCCATA TGGGGATTGG
                GCGCCTTCTC CGTGTACCGA CTTATAGCTG CCAAAGGTAT ACCCCTAACC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5301    TGGCGACGAC TCCTGGAGCC CGTCAGTATC GGCGGAATTC CAGCTGAGCG
                ACCGCTGCTG AGGACCTCGG GCAGTCATAG CCGCCTTAAG GTCGACTCGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5351    CCGGTCGCTA CCATTACCAG TTGGTCTGGT GTCAAAAATA ATAATAACCG
                GGCCAGCGAT GGTAATGGTC AACCAGACCA CAGTTTTTAT TATTATTGGC
        5401    GGCAGGGGGG ATCCGGAGCT TATCGCAGAT CAATTCGATA TCAAGCTTAT
                CCGTCCCCCC TAGGCCTCGA ATAGCGTCTA GTTAAGCTAT AGTTCGAATA
                                                    H6  Promoter
                                                    ~~~~~~~~~~~~~~~~
        5451    CGATACCGTC GACGGTATCG ATAAGCTCTA GTGGAGGGTT CTTTATTCTA
                GCTATGGCAG CTGCCATAGC TATTCGAGAT CACCTCCCAA GAAATAAGAT
                                H6  Promoter
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
```

```
5501   TACTTAAAAA GTGAAAATAA ATACAAAGGT TCTTGAGGGT TGTGTTAAAT
       ATGAATTTTT CACTTTTATT TATGTTTCCA AGAACTCCCA ACACAATTTA
                        H6  Promoter
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5551   TGAAAGCGAG AAATAATCAT AAATTATTTC ATTATCGCGA TATCCGTTAA
       ACTTTCGCTC TTTATTAGTA TTTAATAAAG TAATAGCGCT ATAGGCAATT
        H6  Promoter                          NYESO-1
       ~~~~~~~~~~~~~~~~~                ~~~~~~~~~~~~~~~~~~~~~~~~~~
5601   GTTTGTATCG TACCCCCCCC GAGCCATGCA GGCCGAAGGC CGGGGCACAG
       CAAACATAGC ATGGGGGGGG CTCGGTACGT CCGGCTTCCG GCCCCGTGTC
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5651   GGGGTTCGAC GGGCGATGCT GATGGCCCAG GAGGCCCTGG CATTCCTGAT
       CCCCAAGCTG CCCGCTACGA CTACCGGGTC CTCCGGGACC GTAAGGACTA
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5701   GGCCCAGGGG GCAATGCTGG CGGCCCAGGA GAGGCGGGTG CCACGGGCGG
       CCGGGTCCCC CGTTACGACC GCCGGGTCCT CTCCGCCCAC GGTGCCCGCC
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5751   CAGAGGTCCC CGGGGCGCAG GGGCAGCAAG GGCCTCGGGG CCGGGAGGAG
       GTCTCCAGGG GCCCCGCGTC CCCGTCGTTC CCGGAGCCCC GGCCCTCCTC
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5801   GCGCCCCGCG GGGTCCGCAT GGCGGCGCGG CTTCAGGGCT GAATGGATGC
       CGCGGGGCGC CCCAGGCGTA CCGCCGCGCC GAAGTCCCGA CTTACCTACG
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5851   TGCAGATGCG GGGCCAGGGG GCCGGAGAGC CGCCTGCTTG AGTTCTACCT
       ACGTCTACGC CCCGGTCCCC CGGCCTCTCG GCGGACGAAC TCAAGATGGA
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5901   CGCCATGCCT TTCGCGACAC CCATGGAAGC AGAGCTGGCC CGCAGGAGCC
       GCGGTACGGA AAGCGCTGTG GGTACCTTCG TCTCGACCGG GCGTCCTCGG
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5951   TGGCCCAGGA TGCCCCACCG CTTCCCGTGC CAGGGGTGCT TCTGAAGGAG
       ACCGGGTCCT ACGGGGTGGC GAAGGGCACG GTCCCCACGA AGACTTCCTC
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6001   TTCACTGTGT CCGGCAACAT ACTGACTATC CGACTGACTG CTGCAGACCA
       AAGTGACACA GGCCGTTGTA TGACTGATAG GCTGACTGAC GACGTCTGGT
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6051   CCGCCAACTG CAGCTCTCCA TCAGCTCCTG TCTCCAGCAG CTTTCCCTGT
       GGCGGTTGAC GTCGAGAGGT AGTCGAGGAC AGAGGTCGTC GAAAGGGACA
                           NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6101   TGATGTGGAT CACGCAGGTG TTTCTGCCCG TGTTTTTGGC TCAGCCTCCC
       ACTACACCTA GTGCGTCCAC AAAGACGGGC ACAAAAACCG AGTCGGAGGG
              NYESO-1
       ~~~~~~~~~~~~~~~~~~~~~~~~~~
6151   TCAGGGCAGA GGCGCTAAGT AATTAATTTT TTTTTGGGCT GCAGGATCGC
       AGTCCCGTCT CCGCGATTCA TTAATTAAAA AAAAACCCGA CGTCCTAGCG
```

```
                              sE/L Promoter
                          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6201   TAGCAAAAAT TGAAATTTTA TTTTTTTTTT TTGGAATATA AATAAGCTCG
             ATCGTTTTTA ACTTTAAAAT AAAAAAAAAA AACCTTATAT TTATTCGAGC
                                        hTRP-2
                          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
             sE/L Promoter
             ~~~~
      6251   AAGCTCGAGC CATGAGCCCC CTTTGGTGGG GGTTTCTGCT CAGTTGCTTG
             TTCGAGCTCG GTACTCGGGG GAAACCACCC CCAAAGACGA GTCAACGAAC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6301   GGCTGCAAAA TCCTGCCAGG AGCCCAGGGT CAGTTCCCCC GAGTCTGCAT
             CCGACGTTTT AGGACGGTCC TCGGGTCCCA GTCAAGGGGG CTCAGACGTA
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6351   GACGGTGGAC AGCCTAGTGA ACAAGGAGTG CTGCCCACGC CTGGGTGCAG
             CTGCCACCTG TCGGATCACT TGTTCCTCAC GACGGGTGCG GACCCACGTC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6401   AGTCGGCCAA TGTCTGTGGC TCTCAGCAAG GCCGGGGGCA GTGCACAGAG
             TCAGCCGGTT ACAGACACCG AGAGTCGTTC CGGCCCCCGT CACGTGTCTC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6451   GTGCGAGCCG ACACAAGGCC CTGGAGTGGT CCCTACATCC TACGAAACCA
             CACGCTCGGC TGTGTTCCGG GACCTCACCA GGGATGTAGG ATGCTTTGGT
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6501   GGATGACCGT GAGCTGTGGC CAAGAAAATT CTTCCACCGG ACCTGCAAGT
             CCTACTGGCA CTCGACACCG GTTCTTTTAA GAAGGTGGCC TGGACGTTCA
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6551   GCACAGGAAA CTTTGCCGGC TATAATTGTG GAGACTGCAA GTTTGGCTGG
             CGTGTCCTTT GAAACGGCCG ATATTAACAC CTCTGACGTT CAAACCGACC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6601   ACCGGTCCCA ACTGCGAGCG GAAGAAACCA CCAGTGATTC GGCAGAACAT
             TGGCCAGGGT TGACGCTCGC CTTCTTTGGT GGTCACTAAG CCGTCTTGTA
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6651   CCATTCCTTG AGTCCTCAGG AAAGAGAGCA GTTCTTGGGC GCCTTAGATC
             GGTAAGGAAC TCAGGAGTCC TTTCTCTCGT CAAGAACCCG CGGAATCTAG
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6701   TCGCGAAGAA GAGAGTACAC CCCGACTACG TGATCACCAC ACAACACTGG
             AGCGCTTCTT CTCTCATGTG GGGCTGATGC ACTAGTGGTG TGTTGTGACC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6751   CTGGGCCTGC TTGGGCCCAA TGGAACCCAG CCGCAGTTTG CCAACTGCAG
             GACCCGGACG AACCCGGGTT ACCTTGGGTC GGCGTCAAAC GGTTGACGTC
                                hTRP-2
             ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      6801   TGTTTATGAT TTCTTCGTGT GGCTCCATTA TTATTCTGTT AGAGATACAT
             ACAAATACTA AAGAAGCACA CCGAGGTAAT AATAAGACAA TCTCTATGTA
```

```
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        6851    TATTAGGACC AGGACGCCCC TACAGGGCCA TAGATTTCTC ACATCAAGGA
                ATAATCCTGG TCCTGCGGGG ATGTCCCGGT ATCTAAAGAG TGTAGTTCCT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        6901    CCTGCATTTG TTACCTGGCA CCGGTACCAT TTGTTGTGTC TGGAAAGAGA
                GGACGTAAAC AATGGACCGT GGCCATGGTA AACAACACAG ACCTTTCTCT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        6951    TCTCCAGCGA CTCATTGGCA ATGAGTCTTT TGCTTTGCCC TACTGGAACT
                AGAGGTCGCT GAGTAACCGT TACTCAGAAA ACGAAACGGG ATGACCTTGA
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7001    TTGCCACTGG GAGGAACGAG TGTGATGTGT GTACAGACCA GCTGTTTGGG
                AACGGTGACC CTCCTTGCTC ACACTACACA CATGTCTGGT CGACAAACCC
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7051    GCAGCGAGAC CAGACGATCC GACTCTGATT AGTCGGAACT CAAGATTCTC
                CGTCGCTCTG GTCTGCTAGG CTGAGACTAA TCAGCCTTGA GTTCTAAGAG
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7101    CAGCTGGGAA ACTGTCTGTG ATAGCTTGGA TGACTACAAC CACCTGGTCA
                GTCGACCCTT TGACAGACAC TATCGAACCT ACTGATGTTG GTGGACCAGT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7151    CCTTGTGCAA TGGAACCTAT GAAGGTTTGC TGAGAAGAAA TCAAATGGGA
                GGAACACGTT ACCTTGGATA CTTCCAAACG ACTCTTCTTT AGTTTACCCT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7201    AGAAACAGCA TGAAATTGCC AACCTTAAAA GACATACGAG ATTGCCTGTC
                TCTTTGTCGT ACTTTAACGG TTGGAATTTT CTGTATGCTC TAACGGACAG
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7251    TCTCCAGAAG TTTGACAATC CTCCCTTCTT CCAGAACTCT ACCTTCAGTT
                AGAGGTCTTC AAACTGTTAG GAGGGAAGAA GGTCTTGAGA TGGAAGTCAA
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7301    TCAGGAATGC TTTGGAAGGG TTTGATAAAG CAGATGGGAC TCTGGATTCT
                AGTCCTTACG AAACCTTCCC AAACTATTTC GTCTACCCTG AGACCTAAGA
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7351    CAAGTGATGA GCCTTCATAA TTTGGTTCAT TCCTTCCTGA ACGGGACAAA
                GTTCACTACT CGGAAGTATT AAACCAAGTA AGGAAGGACT TGCCCTGTTT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7401    CGCTTTGCCA CATTCAGCCG CCAATGATCC CATCTTCGTG GTGATTTCTA
                GCGAAACGGT GTAAGTCGGC GGTTACTAGG GTAGAAGCAC CACTAAAGAT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7451    ATCGTTTGCT TTACAATGCT ACAACAAACA TCCTTGAACA TGTAAGAAAA
                TAGCAAACGA AATGTTACGA TGTTGTTTGT AGGAACTTGT ACATTCTTTT
                                hTRP-2
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7501    GAGAAAGCGA CCAAGGAACT CCCTTCCCTG CATGTGCTGG TTCTTCATTC
                CTCTTTCGCT GGTTCCTTGA GGGAAGGGAC GTACACGACC AAGAAGTAAG
```

```
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7551      CTTTACTGAT GCCATCTTTG ATGAGTGGAT GAAAAGATTT AATCCTCCTG
                    GAAATGACTA CGGTAGAAAC TACTCACCTA CTTTTCTAAA TTAGGAGGAC
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7601      CAGATGCCTG GCCTCAGGAG CTGGCCCCTA TTGGTCACAA TCGGATGTAC
                    GTCTACGGAC CGGAGTCCTC GACCGGGGAT AACCAGTGTT AGCCTACATG
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7651      AACATGGTTC CTTTCTTCCC TCCAGTGACT AATGAAGAAC TCTTTTTAAC
                    TTGTACCAAG GAAAGAAGGG AGGTCACTGA TTACTTCTTG AGAAAAATTG
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7701      CTCAGACCAA CTTGGCTACA GCTATGCCAT CGATCTGCCA GTTTCAGTTG
                    GAGTCTGGTT GAACCGATGT CGATACGGTA GCTAGACGGT CAAAGTCAAC
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7751      AAGAAACTCC AGGTTGGCCC ACAACTCTCT TAGTAGTCAT GGGAACACTG
                    TTCTTTGAGG TCCAACCGGG TGTTGAGAGA ATCATCAGTA CCCTTGTGAC
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7801      GTGGCTTTGG TTGGTCTGTT CGTGCTGTTG GCTTTTCTTC AATATAGAAG
                    CACCGAAACC AACCAGACAA GCACGACAAC CGAAAAGAAG TTATATCTTC
                              hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          7851      ACTTCGAAAA GGATATACAC CCCTAATGGA GACACATTTA AGCAGCAAGA
                    TGAAGCTTTT CCTATATGTG GGGATTACCT CTGTGTAAAT TCGTCGTTCT
                           hTRP-2
                    ~~~~~~~~~~~~~~~~~~~~~~
          7901      GATACACAGA AGAAGCCTAG TTTTTTAATT AAGCATGCTC TAGAATCGAT
                    CTATGTGTCT TCTTCGGATC AAAAAATTAA TTCGTACGAG ATCTTAGCTA
                                                        C5 Left Arm
                                                    ~~~~~~~~~~~~~~~~~~~
          7951      CCCGGGTTTT TATGACTAGT TAATCACGGC CGCTTATAAA GATCTAAAAT
                    GGGCCCAAAA ATACTGATCA ATTAGTGCCG GCGAATATTT CTAGATTTTA
                                    C5 Left Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          8001      GCATAATTTC TAAATAATGA AAAAAAAGTA CATCATGAGC AACGCGTTAG
                    CGTATTAAAG ATTTATTACT TTTTTTTCAT GTAGTACTCG TTGCGCAATC
                                    C5 Left Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          8051      TATATTTTAC AATGGAGATT AACGCTCTAT ACCGTTCTAT GTTTATTGAT
                    ATATAAAATG TTACCTCTAA TTGCGAGATA TGGCAAGATA CAAATAACTA
                                    C5 Left Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          8101      TCAGATGATG TTTTAGAAAA GAAAGTTATT GAATATGAAA ACTTTAATGA
                    AGTCTACTAC AAAATCTTTT CTTTCAATAA CTTATACTTT TGAAATTACT
                                    C5 Left Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          8151      AGATGAAGAT GACGACGATG ATTATTGTTG TAAATCTGTT TTAGATGAAG
                    TCTACTTCTA CTGCTGCTAC TAATAACAAC ATTTAGACAA AATCTACTTC
                                    C5 Left Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
          8201      AAGATGACGC GCTAAAGTAT ACTATGGTTA CAAAGTATAA GTCTATACTA
                    TTCTACTGCG CGATTTCATA TGATACCAAT GTTTCATATT CAGATATGAT
```

```
                         C5 Left Arm
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
8251   CTAATGGCGA CTTGTGCAAG AAGGTATAGT ATAGTGAAAA TGTTGTTAGA
       GATTACCGCT GAACACGTTC TTCCATATCA TATCACTTTT ACAACAATCT
                         C5 Left Arm
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
8301   TTATGATTAT GAAAAACCAA ATAAATCAGA TCCATATCTA AAGGTATCTC
       AATACTAATA CTTTTTGGTT TATTTAGTCT AGGTATAGAT TTCCATAGAG
                         C5 Left Arm
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
8351   CTTTGCACAT AATTTCATCT ATTCCTAGTT TAGAATACTT TTCATTATAT
       GAAACGTGTA TTAAAGTAGA TAAGGATCAA ATCTTATGAA AAGTAATATA
                         C5 Left Arm
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
8401   TTGTTTACAG CTGAAGACGA AAAAAATATA TCGATAATAG AAGATTATGT
       AACAAATGTC GACTTCTGCT TTTTTTATAT AGCTATTATC TTCTAATACA
                    C5 Left Arm
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
8451   TAACTCTGCT AATAAGATGA AATTGAATGA GTCTGTGACT GCAGCCAAGC
       ATTGAGACGA TTATTCTACT TTAACTTACT CAGACACTGA CGTCGGTTCG
8501   TTGGCACTGG CCGTCGTTTT ACAACGTCGT GACTGGGAAA ACCCTGGCGT
       AACCGTGACC GGCAGCAAAA TGTTGCAGCA CTGACCCTTT TGGGACCGCA
8551   TACCCAACTT AATCGCCTTG CAGCACATCC CCCTTTCGCC AGCTGGCGTA
       ATGGGTTGAA TTAGCGGAAC GTCGTGTAGG GGGAAAGCGG TCGACCGCAT
8601   ATAGCGAAGA GGCCCGCACC GATCGCCCTT CCCAACAGTT GCGCAGCCTG
       TATCGCTTCT CCGGGCGTGG CTAGCGGGAA GGGTTGTCAA CGCGTCGGAC
8651   AATGGCGAAT GGCGCCTGAT GCGGTATTTT CTCCTTACGC ATCTGTGCGG
       TTACCGCTTA CCGCGGACTA CGCCATAAAA GAGGAATGCG TAGACACGCC
8701   TATTTCACAC CGCATATGGT GCACTCTCAG TACAATCTGC TCTGATGCCG
       ATAAAGTGTG GCGTATACCA CGTGAGAGTC ATGTTAGACG AGACTACGGC
8751   CATAGTTAAG CCAGCCCCGA CACCCGCCAA CACCCGCTGA CGCGCCCTGA
       GTATCAATTC GGTCGGGGCT GTGGGCGGTT GTGGGCGACT GCGCGGGACT
8801   CGGGCTTGTC TGCTCCCGGC ATCCGCTTAC AGACAAGCTG TGACCGTCTC
       GCCCGAACAG ACGAGGGCCG TAGGCGAATG TCTGTTCGAC ACTGGCAGAG
8851   CGGGAGCTGC ATGTGTCAGA GGTTTTCACC GTCATCACCG AAACGCGCGA
       GCCCTCGACG TACACAGTCT CCAAAAGTGG CAGTAGTGGC TTTGCGCGCT
8901   GACGAAAGGG CCTCGTGATA CGCCTATTTT TATAGGTTAA TGTCATGATA
       CTGCTTTCCC GGAGCACTAT GCGGATAAAA ATATCCAATT ACAGTACTAT
8951   ATAATGGTTT CTTAGACGTC AGGTGGCACT TTTCGGGGAA ATGTGCGCGG
       TATTACCAAA GAATCTGCAG TCCACCGTGA AAAGCCCCTT TACACGCGCC
9001   AACCCCTATT TGTTTATTTT TCTAAATACA TTCAAATATG TATCCGCTCA
       TTGGGGATAA ACAAATAAAA AGATTTATGT AAGTTTATAC ATAGGCGAGT
9051   TGAGACAATA ACCCTGATAA ATGCTTCAAT AATATTGAAA AAGGAAGAGT
       ACTCTGTTAT TGGGACTATT TACGAAGTTA TTATAACTTT TTCCTTCTCA
                           Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9101   ATGAGTATTC AACATTTCCG TGTCGCCCTT ATTCCCTTTT TTGCGGCATT
       TACTCATAAG TTGTAAAGGC ACAGCGGGAA TAAGGGAAAA AACGCCGTAA
                           Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9151   TTGCCTTCCT GTTTTTGCTC ACCCAGAAAC GCTGGTGAAA GTAAAAGATG
       AACGGAAGGA CAAAAACGAG TGGGTCTTTG CGACCACTTT CATTTTCTAC
                           Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9201   CTGAAGATCA GTTGGGTGCA CGAGTGGGTT ACATCGAACT GGATCTCAAC
       GACTTCTAGT CAACCCACGT GCTCACCCAA TGTAGCTTGA CCTAGAGTTG
```

```
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9251  AGCGGTAAGA TCCTTGAGAG TTTTCGCCCC GAAGAACGTT TTCCAATGAT
          TCGCCATTCT AGGAACTCTC AAAAGCGGGG CTTCTTGCAA AAGGTTACTA
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9301  GAGCACTTTT AAAGTTCTGC TATGTGGCGC GGTATTATCC CGTATTGACG
          CTCGTGAAAA TTTCAAGACG ATACACCGCG CCATAATAGG GCATAACTGC
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9351  CCGGGCAAGA GCAACTCGGT CGCCGCATAC ACTATTCTCA GAATGACTTG
          GGCCCGTTCT CGTTGAGCCA GCGGCGTATG TGATAAGAGT CTTACTGAAC
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9401  GTTGAGTACT CACCAGTCAC AGAAAAGCAT CTTACGGATG GCATGACAGT
          CAACTCATGA GTGGTCAGTG TCTTTTCGTA GAATGCCTAC CGTACTGTCA
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9451  AAGAGAATTA TGCAGTGCTG CCATAACCAT GAGTGATAAC ACTGCGGCCA
          TTCTCTTAAT ACGTCACGAC GGTATTGGTA CTCACTATTG TGACGCCGGT
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9501  ACTTACTTCT GACAACGATC GGAGGACCGA AGGAGCTAAC CGCTTTTTTG
          TGAATGAAGA CTGTTGCTAG CCTCCTGGCT TCCTCGATTG GCGAAAAAAC
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9551  CACAACATGG GGGATCATGT AACTCGCCTT GATCGTTGGG AACCGGAGCT
          GTGTTGTACC CCCTAGTACA TTGAGCGGAA CTAGCAACCC TTGGCCTCGA
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9601  GAATGAAGCC ATACCAAACG ACGAGCGTGA CACCACGATG CCTGTAGCAA
          CTTACTTCGG TATGGTTTGC TGCTCGCACT GTGGTGCTAC GGACATCGTT
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9651  TGGCAACAAC GTTGCGCAAA CTATTAACTG GCGAACTACT TACTCTAGCT
          ACCGTTGTTG CAACGCGTTT GATAATTGAC CGCTTGATGA ATGAGATCGA
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9701  TCCCGGCAAC AATTAATAGA CTGGATGGAG GCGGATAAAG TTGCAGGACC
          AGGGCCGTTG TTAATTATCT GACCTACCTC CGCCTATTTC AACGTCCTGG
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9751  ACTTCTGCGC TCGGCCCTTC CGGCTGGCTG GTTTATTGCT GATAAATCTG
          TGAAGACGCG AGCCGGGAAG GCCGACCGAC CAAATAACGA CTATTTAGAC
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9801  GAGCCGGTGA GCGTGGGTCT CGCGGTATCA TTGCAGCACT GGGGCCAGAT
          CTCGGCCACT CGCACCCAGA GCGCCATAGT AACGTCGTGA CCCCGGTCTA
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9851  GGTAAGCCCT CCCGTATCGT AGTTATCTAC ACGACGGGGA GTCAGGCAAC
          CCATTCGGGA GGGCATAGCA TCAATAGATG TGCTGCCCCT CAGTCCGTTG
                                Amp(R)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    9901  TATGGATGAA CGAAATAGAC AGATCGCTGA GATAGGTGCC TCACTGATTA
          ATACCTACTT GCTTTATCTG TCTAGCGACT CTATCCACGG AGTGACTAAT
```

Amp(R)
~~~~~~~~~~~~

```
     9951  AGCATTGGTA ACTGTCAGAC CAAGTTTACT CATATATACT TTAGATTGAT
           TCGTAACCAT TGACAGTCTG GTTCAAATGA GTATATATGA AATCTAACTA
    10001  TTAAAACTTC ATTTTTAATT TAAAAGGATC TAGGTGAAGA TCCTTTTTGA
           AATTTTGAAG TAAAAATTAA ATTTTCCTAG ATCCACTTCT AGGAAAAACT
    10051  TAATCTCATG ACCAAAATCC CTTAACGTGA GTTTTCGTTC CACTGAGCGT
           ATTAGAGTAC TGGTTTTAGG GAATTGCACT CAAAAGCAAG GTGACTCGCA
    10101  CAGACCCCGT AGAAAAGATC AAAGGATCTT CTTGAGATCC TTTTTTTCTG
           GTCTGGGGCA TCTTTTCTAG TTTCCTAGAA GAACTCTAGG AAAAAAAGAC
    10151  CGCGTAATCT GCTGCTTGCA AACAAAAAAA CCACCGCTAC CAGCGGTGGT
           GCGCATTAGA CGACGAACGT TTGTTTTTTT GGTGGCGATG GTCGCCACCA
    10201  TTGTTTGCCG GATCAAGAGC TACCAACTCT TTTTCCGAAG GTAACTGGCT
           AACAAACGGC CTAGTTCTCG ATGGTTGAGA AAAAGGCTTC CATTGACCGA
    10251  TCAGCAGAGC GCAGATACCA AATACTGTCC TTCTAGTGTA GCCGTAGTTA
           AGTCGTCTCG CGTCTATGGT TTATGACAGG AAGATCACAT CGGCATCAAT
    10301  GGCCACCACT TCAAGAACTC TGTAGCACCG CCTACATACC TCGCTCTGCT
           CCGGTGGTGA AGTTCTTGAG ACATCGTGGC GGATGTATGG AGCGAGACGA
    10351  AATCCTGTTA CCAGTGGCTG CTGCCAGTGG CGATAAGTCG TGTCTTACCG
           TTAGGACAAT GGTCACCGAC GACGGTCACC GCTATTCAGC ACAGAATGGC
    10401  GGTTGGACTC AAGACGATAG TTACCGGATA AGGCGCAGCG GTCGGGCTGA
           CCAACCTGAG TTCTGCTATC AATGGCCTAT TCCGCGTCGC CAGCCCGACT
    10451  ACGGGGGGTT CGTGCACACA GCCCAGCTTG GAGCGAACGA CCTACACCGA
           TGCCCCCCAA GCACGTGTGT CGGGTCGAAC CTCGCTTGCT GGATGTGGCT
    10501  ACTGAGATAC CTACAGCGTG AGCTATGAGA AAGCGCCACG CTTCCCGAAG
           TGACTCTATG GATGTCGCAC TCGATACTCT TTCGCGGTGC GAAGGGCTTC
    10551  GGAGAAAGGC GGACAGGTAT CCGGTAAGCG GCAGGGTCGG AACAGGAGAG
           CCTCTTTCCG CCTGTCCATA GGCCATTCGC CGTCCCAGCC TTGTCCTCTC
    10601  CGCACGAGGG AGCTTCCAGG GGGAAACGCC TGGTATCTTT ATAGTCCTGT
           GCGTGCTCCC TCGAAGGTCC CCCTTTGCGG ACCATAGAAA TATCAGGACA
    10651  CGGGTTTCGC CACCTCTGAC TTGAGCGTCG ATTTTTGTGA TGCTCGTCAG
           GCCCAAAGCG GTGGAGACTG AACTCGCAGC TAAAAACACT ACGAGCAGTC
    10701  GGGGGCGGAG CCTATGGAAA AACGCCAGCA ACGCGGCCTT TTTACGGTTC
           CCCCCGCCTC GGATACCTTT TTGCGGTCGT TGCGCCGGAA AAATGCCAAG
    10751  CTGGCCTTTT GCTGGCCTTT TGCTCACATG TTCTTTCCTG CGTTATCCCC
           GACCGGAAAA CGACCGGAAA ACGAGTGTAC AAGAAAGGAC GCAATAGGGG
    10801  TGATTCTGTG GATAACCGTA TTACCGCCTT TGAGTGAGCT GATACCGCTC
           ACTAAGACAC CTATTGGCAT AATGGCGGAA ACTCACTCGA CTATGGCGAG
    10851  GCCGCAGCCG AACGACCGAG CGCAGCGAGT CAGTGAGCGA GGAAGCGGAA
           CGGCGTCGGC TTGCTGGCTC GCGTCGCTCA GTCACTCGCT CCTTCGCCTT
    10901  GAGCGCCCAA TACGCAAACC GCCTCTCCCC GCGCGTTGGC CGATTCATTA
           CTCGCGGGTT ATGCGTTTGG CGGAGAGGGG CGCGCAACCG GCTAAGTAAT
    10951  ATGCAGCTGG CACGACAGGT TTCCCGACTG GAAAGCGGGC AGTGAGCGCA
           TACGTCGACC GTGCTGTCCA AAGGGCTGAC CTTTCGCCCG TCACTCGCGT
    11001  ACGCAATTAA TGTGAGTTAG CTCACTCATT AGGCACCCCA GGCTTTACAC
           TGCGTTAATT ACACTCAATC GAGTGAGTAA TCCGTGGGGT CCGAAATGTG
    11051  TTTATGCTTC CGGCTCGTAT GTTGTGTGGA ATTGTGAGCG GATAACAATT
           AAATACGAAG GCCGAGCATA CAACACACCT TAACACTCGC CTATTGTTAA
    11101  TCACACAGGA AACAGCTATG ACCATGATTA CGAATTGAAT TGCGGCCGCA
           AGTGTGTCCT TTGTCGATAC TGGTACTAAT GCTTAACTTA ACGCCGGCGT
    11151  ATTCTAAG
```
~~~~~~~~~~~~

FIGURE 4

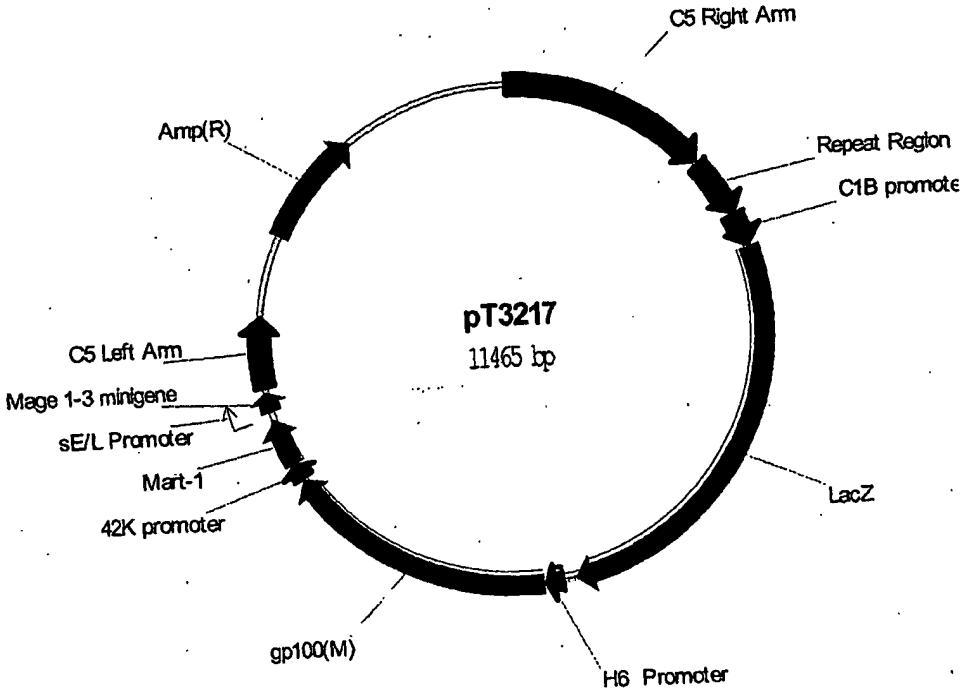

C5 Right Arm
Repeat Region
C1B promote
LacZ
H6 Promoter
gp100(M)
42K promoter
Mart-1
sE/L Promoter
Mage 1-3 minigene
C5 Left Arm
Amp(R)
pT3217
11465 bp

FIGURE 5

DNA Sequence of donor plasmid pT3217

```
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1   TGAATGTTAA ATGTTATACT TTGGATGAAG CTATAAATAT GCATTGGAAA
          ACTTACAATT TACAATATGA AACCTACTTC GATATTTATA CGTAACCTTT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     51   AATAATCCAT TTAAAGAAAG GATTCAAATA CTACAAAACC TAAGCGATAA
          TTATTAGGTA AATTTCTTTC CTAAGTTTAT GATGTTTTGG ATTCGCTATT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    101   TATGTTAACT AAGCTTATTC TTAACGACGC TTTAAATATA CACAAATAAA
          ATACAATTGA TTCGAATAAG AATTGCTGCG AAATTTATAT GTGTTTATTT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    151   CATAATTTTT GTATAACCTA ACAAATAACT AAAACATAAA AATAATAAAA
          GTATTAAAAA CATATTGGAT TGTTTATTGA TTTTGTATTT TTATTATTTT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    201   GGAAATGTAA TATCGTAATT ATTTTACTCA GGAATGGGGT TAAATATTTA
          CCTTTACATT ATAGCATTAA TAAAATGAGT CCTTACCCCA ATTTATAAAT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    251   TATCACGTGT ATATCTATAC TGTTATCGTA TACTCTTTAC AATTACTATT
          ATAGTGCACA TATAGATATG ACAATAGCAT ATGAGAAATG TTAATGATAA
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    301   ACGAATATGC AAGAGATAAT AAGATTACGT ATTTAAGAGA ATCTTGTCAT
          TGCTTATACG TTCTCTATTA TTCTAATGCA TAAATTCTCT TAGAACAGTA
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    351   GATAATTGGG TACGACATAG TGATAAATGC TATTTCGCAT CGTTACATAA
          CTATTAACCC ATGCTGTATC ACTATTTACG ATAAAGCGTA GCAATGTATT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    401   AGTCAGTTGG AAAGATGGAT TTGACAGATG TAACTTAATA GGTGCAAAAA
          TCAGTCAACC TTTCTACCTA AACTGTCTAC ATTGAATTAT CCACGTTTTT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    451   TGTTAAATAA CAGCATTCTA TCGGAAGATA GGATACCAGT TATATTATAC
          ACAATTTATT GTCGTAAGAT AGCCTTCTAT CCTATGGTCA ATATAATATG
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    501   AAAAATCACT GGTTGGATAA AACAGATTCT GCAATATTCG TAAAAGATGA
          TTTTTAGTGA CCAACCTATT TTGTCTAAGA CGTTATAAGC ATTTTCTACT
                        C5 Right Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    551   AGATTACTGC GAATTTGTAA ACTATGACAA TAAAAAGCCA TTTATCTCAA
          TCTAATGACG CTTAAACATT TGATACTGTT ATTTTTCGGT AAATAGAGTT
```

```
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     601    CGACATCGTG TAATTCTTCC ATGTTTTATG TATGTGTTTC AGATATTATG
            GCTGTAGCAC ATTAAGAAGG TACAAAATAC ATACACAAAG TCTATAATAC
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     651    AGATTACTAT AAACTTTTTG TATACTTATA TTCCGTAAAC TATATTAATC
            TCTAATGATA TTTGAAAAAC ATATGAATAT AAGGCATTTG ATATAATTAG
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     701    ATGAAGAAAA TGAAAAAGTA TAGAAGCTGT TCACGAGCGG TTGTTGAAAA
            TACTTCTTTT ACTTTTTCAT ATCTTCGACA AGTGCTCGCC AACAACTTTT
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     751    CAACAAAATT ATACATTCAA GATGGCTTAC ATATACGTCT GTGAGGCTAT
            GTTGTTTTAA TATGTAAGTT CTACCGAATG TATATGCAGA CACTCCGATA
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     801    CATGGATAAT GACAATGCAT CTCTAAATAG GTTTTTGGAC AATGGATTCG
            GTACCTATTA CTGTTACGTA GAGATTTATC CAAAAACCTG TTACCTAAGC
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     851    ACCCTAACAC GGAATATGGT ACTCTACAAT CTCCTCTTGA AATGGCTGTA
            TGGGATTGTG CCTTATACCA TGAGATGTTA GAGGAGAACT TTACCGACAT
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     901    ATGTTCAAGA ATACCGAGGC TATAAAAATC TTGATGAGGT ATGGAGCTAA
            TACAAGTTCT TATGGCTCCG ATATTTTTAG AACTACTCCA TACCTCGATT
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     951    ACCTGTAGTT ACTGAATGCA CAACTTCTTG TCTGCATGAT GCGGTGTTGA
            TGGACATCAA TGACTTACGT GTTGAAGAAC AGACGTACTA CGCCACAACT
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1001    GAGACGACTA CAAAATAGTG AAAGATCTGT TGAAGAATAA CTATGTAAAC
            CTCTGCTGAT GTTTTATCAC TTTCTAGACA ACTTCTTATT GATACATTTG
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1051    AATGTTCTTT ACAGCGGAGG CTTTACTCCT TTGTGTTTGG CAGCTTACCT
            TTACAAGAAA TGTCGCCTCC GAAATGAGGA AACACAAACC GTCGAATGGA
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1101    TAACAAAGTT AATTTGGTTA AACTTCTATT GGCTCATTCG GCGGATGTAG
            ATTGTTTCAA TTAAACCAAT TTGAAGATAA CCGAGTAAGC CGCCTACATC
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1151    ATATTTCAAA CACGGATCGG TTAACTCCTC TACATATAGC CGTATCAAAT
            TATAAAGTTT GTGCCTAGCC AATTGAGGAG ATGTATATCG GCATAGTTTA
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1201    AAAAATTTAA CAATGGTTAA ACTTCTATTG AACAAAGGTG CTGATACTGA
            TTTTTAAATT GTTACCAATT TGAAGATAAC TTGTTTCCAC GACTATGACT
                          C5 Right Arm
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    1251    CTTGCTGGAT AACATGGGAT GTACTCCTTT AATGATCGCT GTACAATCTG
            GAACGACCTA TTGTACCCTA CATGAGGAAA TTACTAGCGA CATGTTAGAC
```

```
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1301      GAAATATTGA AATATGTAGC ACACTACTTA AAAAAAATAA AATGTCCAGA
                CTTTATAACT TTATACATCG TGTGATGAAT TTTTTTTATT TTACAGGTCT
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1351      ACTGGGAAAA ATTGATCTTG CCAGCTGTAA TTCATGGTAG AAAAGAAGTG
                TGACCCTTTT TAACTAGAAC GGTCGACATT AAGTACCATC TTTTCTTCAC
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1401      CTCAGGCTAC TTTTCAACAA AGGAGCAGAT GTAAACTACA TCTTTGAAAG
                GAGTCCGATG AAAAGTTGTT TCCTCGTCTA CATTTGATGT AGAAACTTTC
                          C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1451      AAATGGAAAA TCATATACTG TTTTGGAATT GATTAAAGAA AGTTACTCTG
                TTTACCTTTT AGTATATGAC AAAACCTTAA CTAATTTCTT TCAATGAGAC
                           C5 Right Arm
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1501      AGACACAAAA GAGGTAGCTG AAGTGGTACT CTCAAAGGTA CGTGACTAAT
                TCTGTGTTTT CTCCATCGAC TTCACCATGA GAGTTTCCAT GCACTGATTA
                                          Repeat Region
                                        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1551      TAGCTATAAA AAGGATCGGG TTCTTTATTC TATACTTAAA AAGTGAAAAT
                ATCGATATTT TTCCTAGCCC AAGAAATAAG ATATGAATTT TTCACTTTTA
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1601      AAATACAAAG GTTCTTGAGG GTTGTGTTAA ATTGAAAGCG AGAAATAATC
                TTTATGTTTC CAAGAACTCC CAACACAATT TAACTTTCGC TCTTTATTAG
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1651      ATAAATTATT TCATTATCGC GATATCCGTT AAGTTTGTAT CGTAATCTGC
                TATTTAATAA AGTAATAGCG CTATAGGCAA TTCAAACATA GCATTAGACG
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1701      AGCCCCCACC ATGGATCTGG TGCTAAAAAG ATGCCTTCTT CATTTGGCTG
                TCGGGGGTGG TACCTAGACC ACGATTTTTC TACGGAAGAA GTAAACCGAC
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1751      TGATAGGTGC TTTGCTGGCT GTGGGGGCTA CAAAAGTACC CAGAAACCAG
                ACTATCCACG AAACGACCGA CACCCCCGAT GTTTTCATGG GTCTTTGGTC
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1801      GACTGGCTTG GTGTCTCAAG GCAACTCAGA ACCAAAGCCT GGAACAGGCA
                CTGACCGAAC CACAGAGTTC CGTTGAGTCT TGGTTTCGGA CCTTGTCCGT
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1851      GCTGTATCCA GAGTGGACAG AAGCCCAGAG ACTTGACTGC TGGAGAGGTG
                CGACATAGGT CTCACCTGTC TTCGGGTCTC TGAACTGACG ACCTCTCCAC
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1901      GTCAAGTGTC CCTCAAGGTC AGTAATGATG GGCCTACACT GATTGGTGCA
                CAGTTCACAG GGAGTTCCAG TCATTACTAC CCGGATGTGA CTAACCACGT
                          Repeat Region
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      1951      AATGCCTCCT TCTCTATTGC CTTGAACTTC CCTGGAAGCC AAAAGGTATT
                TTACGGAGGA AGAGATAACG GAACTTGAAG GGACCTTCGG TTTTCCATAA
```

```
          Repeat Region                    C1B promoter
          ~~~~~~~                          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2001  GCCAGATACT AGTTCTAGAG GATCATTATT TAACGTAAAC TAAATGGAAA
          CGGTCTATGA TCAAGATCTC CTAGTAATAA ATTGCATTTG ATTTACCTTT
                              C1B promoter
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2051  AGCTATTTAC AGGTACATAC GGTGTTTTTC TGGAATCAAA TGATTCTGAT
          TCGATAAATG TCCATGTATG CCACAAAAAG ACCTTAGTTT ACTAAGACTA
                              C1B promoter
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2101  TTTGAGGATT TTATCAATAC AATAATGACA GTGCTAACTG GTAAAAAAGA
          AAACTCCTAA AATAGTTATG TTATTACTGT CACGATTGAC CATTTTTTCT
                              C1B promoter
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2151  AAGCAAACAA TTATCATGGC TAACAATTTT TATTATATTT GTAGTATGCA
          TTCGTTTGTT AATAGTACCG ATTGTTAAAA ATAATATAAA CATCATACGT
                              C1B promoter
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2201  TAGTGGTCTT TACGTTTCTT TATTTAAAGT TAATGTGTTA AGATTAAATG
          ATCACCAGAA ATGCAAAGAA ATAAATTTCA ATTACACAAT TCTAATTTAC
          C1B promoter                       LacZ
          ~~~~~~~~~~~~~~~~                   ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2251  GAGTAATTGG ATCCCCCATC GATGGGGAAT TCACTGGCCG TCGTTTTACA
          CTCATTAACC TAGGGGGTAG CTACCCCTTA AGTGACCGGC AGCAAAATGT
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2301  ACGTCGTGAC TGGGAAAACC CTGGCGTTAC CCAACTTAAT CGCCTTGCAG
          TGCAGCACTG ACCCTTTTGG GACCGCAATG GGTTGAATTA GCGGAACGTC
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2351  CACATCCCCC TTTCGCCAGC TGGCGTAATA GCGAAGAGGC CCGCACCGAT
          GTGTAGGGGG AAAGCGGTCG ACCGCATTAT CGCTTCTCCG GGCGTGGCTA
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2401  CGCCCTTCCC AACAGTTGCG CAGCCTGAAT GGCGAATGGC GCTTTGCCTG
          GCGGGAAGGG TTGTCAACGC GTCGGACTTA CCGCTTACCG CGAAACGGAC
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2451  GTTTCCGGCA CCAGAAGCGG TGCCGGAAAG CTGGCTGGAG TGCGATCTTC
          CAAAGGCCGT GGTCTTCGCC ACGGCCTTTC GACCGACCTC ACGCTAGAAG
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2501  CTGAGGCCGA TACTGTCGTC GTCCCCTCAA ACTGGCAGAT GCACGGTTAC
          GACTCCGGCT ATGACAGCAG CAGGGGAGTT TGACCGTCTA CGTGCCAATG
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2551  GATGCGCCCA TCTACACCAA CGTGACCTAT CCCATTACGG TCAATCCGCC
          CTACGCGGGT AGATGTGGTT GCACTGGATA GGGTAATGCC AGTTAGGCGG
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2601  GTTTGTTCCC ACGGAGAATC CGACGGGTTG TTACTCGCTC ACATTTAATG
          CAAACAAGGG TGCCTCTTAG GCTGCCCAAC AATGAGCGAG TGTAAATTAC
                                    LacZ
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    2651  TTGATGAAAG CTGGCTACAG GAAGGCCAGA CGCGAATTAT TTTTGATGGC
          AACTACTTTC GACCGATGTC CTTCCGGTCT GCGCTTAATA AAAACTACCG
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2701      GTTAACTCGG CGTTTCATCT GTGGTGCAAC GGGCGCTGGG TCGGTTACGG
                CAATTGAGCC GCAAAGTAGA CACCACGTTG CCCGCGACCC AGCCAATGCC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2751      CCAGGACAGT CGTTTGCCGT CTGAATTTGA CCTGAGCGCA TTTTTACGCG
                GGTCCTGTCA GCAAACGGCA GACTTAAACT GGACTCGCGT AAAAATGCGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2801      CCGGAGAAAA CCGCCTCGCG GTGATGGTGC TGCGCTGGAG TGACGGCAGT
                GGCCTCTTTT GGCGGAGCGC CACTACCACG ACGCGACCTC ACTGCCGTCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2851      TATCTGGAAG ATCAGGATAT GTGGCGGATG AGCGGCATTT TCCGTGACGT
                ATAGACCTTC TAGTCCTATA CACCGCCTAC TCGCCGTAAA AGGCACTGCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2901      CTCGTTGCTG CATAAACCGA CTACACAAAT CAGCGATTTC CATGTTGCCA
                GAGCAACGAC GTATTTGGCT GATGTGTTTA GTCGCTAAAG GTACAACGGT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      2951      CTCGCTTTAA TGATGATTTC AGCCGCGCTG TACTGGAGGC TGAAGTTCAG
                GAGCGAAATT ACTACTAAAG TCGGCGCGAC ATGACCTCCG ACTTCAAGTC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3001      ATGTGCGGCG AGTTGCGTGA CTACCTACGG GTAACAGTTT CTTTATGGCA
                TACACGCCGC TCAACGCACT GATGGATGCC CATTGTCAAA GAAATACCGT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3051      GGGTGAAACG CAGGTCGCCA GCGGCACCGC GCCTTTCGGC GGTGAAATTA
                CCCACTTTGC GTCCAGCGGT CGCCGTGGCG CGGAAAGCCG CCACTTTAAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3101      TCGATGAGCG TGGTGGTTAT GCCGATCGCG TCACACTACG TCTGAACGTC
                AGCTACTCGC ACCACCAATA CGGCTAGCGC AGTGTGATGC AGACTTGCAG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3151      GAAAACCCGA AACTGTGGAG CGCCGAAATC CCGAATCTCT ATCGTGCGGT
                CTTTTGGGCT TTGACACCTC GCGGCTTTAG GGCTTAGAGA TAGCACGCCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3201      GGTTGAACTG CACACCGCCG ACGGCACGCT GATTGAAGCA GAAGCCTGCG
                CCAACTTGAC GTGTGGCGGC TGCCGTGCGA CTAACTTCGT CTTCGGACGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3251      ATGTCGGTTT CCGCGAGGTG CGGATTGAAA ATGGTCTGCT GCTGCTGAAC
                TACAGCCAAA GGCGCTCCAC GCCTAACTTT TACCAGACGA CGACGACTTG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3301      GGCAAGCCGT TGCTGATTCG AGGCGTTAAC CGTCACGAGC ATCATCCTCT
                CCGTTCGGCA ACGACTAAGC TCCGCAATTG GCAGTGCTCG TAGTAGGAGA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3351      GCATGGTCAG GTCATGGATG AGCAGACGAT GGTGCAGGAT ATCCTGCTGA
                CGTACCAGTC CAGTACCTAC TCGTCTGCTA CCACGTCCTA TAGGACGACT
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3401      TGAAGCAGAA CAACTTTAAC GCCGTGCGCT GTTCGCATTA TCCGAACCAT
                ACTTCGTCTT GTTGAAATTG CGGCACGCGA CAAGCGTAAT AGGCTTGGTA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3451      CCGCTGTGGT ACACGCTGTG CGACCGCTAC GGCCTGTATG TGGTGGATGA
                GGCGACACCA TGTGCGACAC GCTGGCGATG CCGGACATAC ACCACCTACT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3501      AGCCAATATT GAAACCCACG GCATGGTGCC AATGAATCGT CTGACCGATG
                TCGGTTATAA CTTTGGGTGC CGTACCACGG TTACTTAGCA GACTGGCTAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3551      ATCCGCGCTG GCTACCGGCG ATGAGCGAAC GCGTAACGCG AATGGTGCAG
                TAGGCGCGAC CGATGGCCGC TACTCGCTTG CGCATTGCGC TTACCACGTC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3601      CGCGATCGTA ATCACCCGAG TGTGATCATC TGGTCGCTGG GGAATGAATC
                GCGCTAGCAT TAGTGGGCTC ACACTAGTAG ACCAGCGACC CCTTACTTAG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3651      AGGCCACGGC GCTAATCACG ACGCGCTGTA TCGCTGGATC AAATCTGTCG
                TCCGGTGCCG CGATTAGTGC TGCGCGACAT AGCGACCTAG TTTAGACAGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3701      ATCCTTCCCG CCCGGTGCAG TATGAAGGCG GCGGAGCCGA CACCACGGCC
                TAGGAAGGGC GGGCCACGTC ATACTTCCGC CGCCTCGGCT GTGGTGCCGG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3751      ACCGATATTA TTTGCCCGAT GTACGCGCGC GTGGATGAAG ACCAGCCCTT
                TGGCTATAAT AAACGGGCTA CATGCGCGCG CACCTACTTC TGGTCGGGAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3801      CCCGGCTGTG CCGAAATGGT CCATCAAAAA ATGGCTTTCG CTACCTGGAG
                GGGCCGACAC GGCTTTACCA GGTAGTTTTT TACCGAAAGC GATGGACCTC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3851      AGACGCGCCC GCTGATCCTT TGCGAATACG CCCACGCGAT GGGTAACAGT
                TCTGCGCGGG CGACTAGGAA ACGCTTATGC GGGTGCGCTA CCCATTGTCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3901      CTTGGCGGTT TCGCTAAATA CTGGCAGGCG TTTCGTCAGT ATCCCCGTTT
                GAACCGCCAA AGCGATTTAT GACCGTCCGC AAAGCAGTCA TAGGGGCAAA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      3951      ACAGGGCGGC TTCGTCTGGG ACTGGGTGGA TCAGTCGCTG ATTAAATATG
                TGTCCCGCCG AAGCAGACCC TGACCCACCT AGTCAGCGAC TAATTTATAC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4001      ATGAAAACGG CAACCCGTGG TCGGCTTACG GCGGTGATTT TGGCGATACG
                TACTTTTGCC GTTGGGCACC AGCCGAATGC CGCCACTAAA ACCGCTATGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      4051      CCGAACGATC GCCAGTTCTG TATGAACGGT CTGGTCTTTG CCGACCGCAC
                GGCTTGCTAG CGGTCAAGAC ATACTTGCCA GACCAGAAAC GGCTGGCGTG
```

```
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4101    GCCGCATCCA GCGCTGACGG AAGCAAAACA CCAGCAGCAG TTTTTCCAGT
                CGGCGTAGGT CGCGACTGCC TTCGTTTTGT GGTCGTCGTC AAAAAGGTCA
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4151    TCCGTTTATC CGGGCAAACC ATCGAAGTGA CCAGCGAATA CCTGTTCCGT
                AGGCAAATAG GCCCGTTTGG TAGCTTCACT GGTCGCTTAT GGACAAGGCA
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4201    CATAGCGATA ACGAGCTCCT GCACTGGATG GTGGCGCTGG ATGGTAAGCC
                GTATCGCTAT TGCTCGAGGA CGTGACCTAC CACCGCGACC TACCATTCGG
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4251    GCTGGCAAGC GGTGAAGTGC CTCTGGATGT CGCTCCACAA GGTAAACAGT
                CGACCGTTCG CCACTTCACG GAGACCTACA GCGAGGTGTT CCATTTGTCA
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4301    TGATTGAACT GCCTGAACTA CCGCAGCCGG AGAGCGCCGG GCAACTCTGG
                ACTAACTTGA CGGACTTGAT GGCGTCGGCC TCTCGCGGCC CGTTGAGACC
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4351    CTCACAGTAC GCGTAGTGCA ACCGAACGCG ACCGCATGGT CAGAAGCCGG
                GAGTGTCATG CGCATCACGT TGGCTTGCGC TGGCGTACCA GTCTTCGGCC
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4401    GCACATCAGC GCCTGGCAGC AGTGGCGTCT GGCGGAAAAC CTCAGTGTGA
                CGTGTAGTCG CGGACCGTCG TCACCGCAGA CCGCCTTTTG GAGTCACACT
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4451    CGCTCCCCGC CGCGTCCCAC GCCATCCCGC ATCTGACCAC CAGCGAAATG
                GCGAGGGGCG GCGCAGGGTG CGGTAGGGCG TAGACTGGTG GTCGCTTTAC
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4501    GATTTTTGCA TCGAGCTGGG TAATAAGCGT TGGCAATTTA ACCGCCAGTC
                CTAAAAACGT AGCTCGACCC ATTATTCGCA ACCGTTAAAT TGGCGGTCAG
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4551    AGGCTTTCTT TCACAGATGT GGATTGGCGA TAAAAAACAA CTGCTGACGC
                TCCGAAAGAA AGTGTCTACA CCTAACCGCT ATTTTTTGTT GACGACTGCG
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4601    CGCTGCGCGA TCAGTTCACC CGTGCACCGC TGGATAACGA CATTGGCGTA
                GCGACGCGCT AGTCAAGTGG GCACGTGGCG ACCTATTGCT GTAACCGCAT
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4651    AGTGAAGCGA CCCGCATTGA CCCTAACGCC TGGGTCGAAC GCTGGAAGGC
                TCACTTCGCT GGGCGTAACT GGGATTGCGG ACCCAGCTTG CGACCTTCCG
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4701    GGCGGGCCAT TACCAGGCCG AAGCAGCGTT GTTGCAGTGC ACGGCAGATA
                CCGCCCGGTA ATGGTCCGGC TTCGTCGCAA CAACGTCACG TGCCGTCTAT
                              LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4751    CACTTGCTGA TGCGGTGCTG ATTACGACCG CTCACGCGTG GCAGCATCAG
                GTGAACGACT ACGCCACGAC TAATGCTGGC GAGTGCGCAC CGTCGTAGTC
```

```
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4801    GGGAAAACCT TATTTATCAG CCGGAAAACC TACCGGATTG ATGGTAGTGG
                CCCTTTTGGA ATAAATAGTC GGCCTTTTGG ATGGCCTAAC TACCATCACC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4851    TCAAATGGCG ATTACCGTTG ATGTTGAAGT GGCGAGCGAT ACACCGCATC
                AGTTTACCGC TAATGGCAAC TACAACTTCA CCGCTCGCTA TGTGGCGTAG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4901    CGGCGCGGAT TGGCCTGAAC TGCCAGCTGG CGCAGGTAGC AGAGCGGGTA
                GCCGCGCCTA ACCGGACTTG ACGGTCGACC GCGTCCATCG TCTCGCCCAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        4951    AACTGGCTCG GATTAGGGCC GCAAGAAAAC TATCCCGACC GCCTTACTGC
                TTGACCGAGC CTAATCCCGG CGTTCTTTTG ATAGGGCTGG CGGAATGACG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5001    CGCCTGTTTT GACCGCTGGG ATCTGCCATT GTCAGACATG TATACCCCGT
                GCGGACAAAA CTGGCGACCC TAGACGGTAA CAGTCTGTAC ATATGGGGCA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5051    ACGTCTTCCC GAGCGAAAAC GGTCTGCGCT GCGGGACGCG CGAATTGAAT
                TGCAGAAGGG CTCGCTTTTG CCAGACGCGA CGCCCTGCGC GCTTAACTTA
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5101    TATGGCCCAC ACCAGTGGCG CGGCGACTTC CAGTTCAACA TCAGCCGGTA
                ATACCGGGTG TGGTCACCGC GCCGCTGAAG GTCAAGTTGT AGTCGGCCAT
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5151    CAGTCAACAG CAATTGATGG AAACCAGCCA TTCGCCATCT GCTGCACGCG
                GTCAGTTGTC GTTAACTACC TTTGGTCGGT AAGCGGTAGA CGACGTGCGC
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5201    GAAGAGGCAC ATGGCTGAAT ATCGACGGTT TCCATATGGG GATTGGTGGC
                CTTCTCCGTG TACCGACTTA TAGCTGCCAA AGGTATACCC CTAACCACCG
                                LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5251    GACGACTCCT GGAGCCCGTC AGTATCGGCG GAATTCCAGC TGAGCGCCGG
                CTGCTGAGGA CCTCGGGCAG TCATAGCCGC CTTAAGGTCG ACTCGCGGCC
                          LacZ
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5301    TCGCTACCAT TACCAGTTGG TCTGGTGTCA AAAATAATAA TAACCGGGCA
                AGCGATGGTA ATGGTCAACC AGACCACAGT TTTTATTATT ATTGGCCCGT
        5351    GGGGGGATCC GGAGCTTATC GCAGATCAAT TCGATATCAA GCTTATCGAT
                CCCCCCTAGG CCTCGAATAG CGTCTAGTTA AGCTATAGTT CGAATAGCTA
                                                  H6  Promoter
                                                  ~~~~~~~~~~~~~~~~~~
        5401    ACCGTCGACC TCGAGTCTAG AATCGATCCC GGGTTCTTTA TTCTATACTT
                TGGCAGCTGG AGCTCAGATC TTAGCTAGGG CCCAAGAAAT AAGATATGAA
                               H6  Promoter
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        5451    AAAAAGTGAA AATAAATACA AAGGTTCTTG AGGGTTGTGT TAAATTGAAA
                TTTTTCACTT TTATTTATGT TTCCAAGAAC TCCCAACACA ATTTAACTTT
```

```
          H6  Promoter
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5501      GCGAGAAATA ATCATAAATT ATTTCATTAT CGCGATATCC GTTAAGTTTG
          CGCTCTTTAT TAGTATTTAA TAAAGTAATA GCGCTATAGG CAATTCAAAC
          H6  Promoter                          gp100(M)
          ~~~~~~~                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5551      TATCGTAATC TGCAGCCCCC ACCATGGATC TGGTGCTAAA AAGATGCCTT
          ATAGCATTAG ACGTCGGGGG TGGTACCTAG ACCACGATTT TTCTACGGAA
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5601      CTTCATTTGG CTGTGATAGG TGCTTTGCTG GCTGTGGGGG CTACAAAAGT
          GAAGTAAACC GACACTATCC ACGAAACGAC CGACACCCCC GATGTTTTCA
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5651      ACCCAGAAAC CAGGACTGGC TTGGTGTCTC AAGGCAACTC AGAACCAAAG
          TGGGTCTTTG GTCCTGACCG AACCACAGAG TTCCGTTGAG TCTTGGTTTC
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5701      CCTGGAACAG GCAGCTGTAT CCAGAGTGGA CAGAAGCCCA GAGACTTGAC
          GGACCTTGTC CGTCGACATA GGTCTCACCT GTCTTCGGGT CTCTGAACTG
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5751      TGCTGGAGAG GTGGTCAAGT GTCCCTCAAG GTCAGTAATG ATGGGCCTAC
          ACGACCTCTC CACCAGTTCA CAGGGAGTTC CAGTCATTAC TACCCGGATG
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5801      ACTGATTGGT GCAAATGCCT CCTTCTCTAT TGCCTTGAAC TTCCCTGGAA
          TGACTAACCA CGTTTACGGA GGAAGAGATA ACGGAACTTG AAGGGACCTT
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5851      GCCAAAAGGT ATTGCCAGAT GGGCAGGTTA TCTGGGTCAA CAATACCATC
          CGGTTTTCCA TAACGGTCTA CCCGTCCAAT AGACCCAGTT GTTATGGTAG
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5901      ATCAATGGGA GCCAGGTGTG GGGAGGACAG CCAGTGTATC CCCAGGAAAC
          TAGTTACCCT CGGTCCACAC CCCTCCTGTC GGTCACATAG GGGTCCTTTG
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
5951      TGACGATGCC TGCATCTTCC CTGATGGTGG ACCTTGCCCA TCTGGCTCTT
          ACTGCTACGG ACGTAGAAGG GACTACCACC TGGAACGGGT AGACCGAGAA
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6001      GGTCTCAGAA GAGAAGCTTT GTTTATGTCT GGAAGACCTG GGGCCAATAC
          CCAGAGTCTT CTCTTCGAAA CAAATACAGA CCTTCTGGAC CCCGGTTATG
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6051      TGGCAAGTTC TAGGGGGCCC AGTGTCTGGG CTGAGCATTG GGACAGGCAG
          ACCGTTCAAG ATCCCCCGGG TCACAGACCC GACTCGTAAC CCTGTCCGTC
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6101      GGCAATGCTG GGCACACACA CGATGGAAGT GACTGTCTAC CATCGCCGGG
          CCGTTACGAC CCGTGTGTGT GCTACCTTCA CTGACAGATG GTAGCGGCCC
                                gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
6151      GATCCCGGAG CTATGTGCCT CTTGCTCATT CCAGCTCAGC CTTCACCATT
          CTAGGGCCTC GATACACGGA GAACGAGTAA GGTCGAGTCG GAAGTGGTAA
```

```
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6201  ATGGACCAGG TGCCTTTCTC CGTGAGCGTG TCCCAGTTGC GGGCCTTGGA
          TACCTGGTCC ACGGAAAGAG GCACTCGCAC AGGGTCAACG CCCGGAACCT
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6251  TGGAGGGAAC AAGCACTTCC TGAGAAATCA GCCTCTGACC TTTGCCCTCC
          ACCTCCCTTG TTCGTGAAGG ACTCTTTAGT CGGAGACTGG AAACGGGAGG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6301  AGCTCCATGA CCCCAGTGGC TATCTGGCTG AAGCTGACCT CTCCTACACC
          TCGAGGTACT GGGGTCACCG ATAGACCGAC TTCGACTGGA GAGGATGTGG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6351  TGGGACTTTG GAGACAGTAG TGGAACCCTG ATCTCTCGGG CACTTGTGGT
          ACCCTGAAAC CTCTGTCATC ACCTTGGGAC TAGAGAGCCC GTGAACACCA
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6401  CACTCATACT TACCTGGAGC CTGGCCCAGT CACTGTTCAG GTGGTCCTGC
          GTGAGTATGA ATGGACCTCG GACCGGGTCA GTGACAAGTC CACCAGGACG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6451  AGGCTGCCAT TCCTCTCACC TCCTGTGGCT CCTCCCCAGT TCCAGGCACC
          TCCGACGGTA AGGAGAGTGG AGGACACCGA GGAGGGGTCA AGGTCCGTGG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6501  ACAGATGGGC ACAGGCCAAC TGCAGAGGCC CCTAACACCA CAGCTGGCCA
          TGTCTACCCG TGTCCGGTTG ACGTCTCCGG GGATTGTGGT GTCGACCGGT
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6551  AGTGCCTACT ACAGAAGTTG TGGGTACTAC ACCTGGTCAG GCGCCAACTG
          TCACGGATGA TGTCTTCAAC ACCCATGATG TGGACCAGTC CGCGGTTGAC
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6601  CAGAGCCCTC TGGAACCACA TCTGTGCAGG TGCCAACCAC TGAAGTCATA
          GTCTCGGGAG ACCTTGGTGT AGACACGTCC ACGGTTGGTG ACTTCAGTAT
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6651  AGCACTGCAC CTGTGCAGAT GCCAACTGCA GAGAGCACAG GTATGACACC
          TCGTGACGTG GACACGTCTA CGGTTGACGT CTCTCGTGTC CATACTGTGG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6701  TGAGAAGGTG CCAGTTTCAG AGGTCATGGG TACCACACTG GCAGAGATGT
          ACTCTTCCAC GGTCAAAGTC TCCAGTACCC ATGGTGTGAC CGTCTCTACA
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6751  CAACTCCAGA GGCTACAGGT ATGACACCTG CAGAGGTATC AATTGTGGTG
          GTTGAGGTCT CCGATGTCCA TACTGTGGAC GTCTCCATAG TTAACACCAC
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6801  CTTTCTGGAA CCACAGCTGC ACAGGTAACA ACTACAGAGT GGGTGGAGAC
          GAAAGACCTT GGTGTCGACG TGTCCATTGT TGATGTCTCA CCCACCTCTG
                              gp100(M)
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6851  CACAGCTAGA GAGCTACCTA TCCCTGAGCC TGAAGGTCCA GATGCCAGCT
          GTGTCGATCT CTCGATGGAT AGGGACTCGG ACTTCCAGGT CTACGGTCGA
```

```
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6901    CAATCATGTC TACGGAAAGT ATTACAGGTT CCCTGGGCCC CCTGCTGGAT
            GTTAGTACAG ATGCCTTTCA TAATGTCCAA GGGACCCGGG GGACGACCTA
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    6951    GGTACAGCCA CCTTAAGGCT GGTGAAGAGA CAAGTCCCCC TGGATTGTGT
            CCATGTCGGT GGAATTCCGA CCACTTCTCT GTTCAGGGGG ACCTAACACA
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7001    TCTGTATCGA TATGGTTCCT TTTCCGTCAC CCTGGACATT GTCCAGGGTA
            AGACATAGCT ATACCAAGGA AAAGGCAGTG GGACCTGTAA CAGGTCCCAT
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7051    TTGAAAGTGC CGAGATCCTG CAGGCTGTGC CGTCCGGTGA GGGGGATGCA
            AACTTTCACG GCTCTAGGAC GTCCGACACG GCAGGCCACT CCCCCTACGT
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7101    TTTGAGCTGA CTGTGTCCTG CCAAGGCGGG CTGCCCAAGG AAGCCTGCAT
            AAACTCGACT GACACAGGAC GGTTCCGCCC GACGGGTTCC TTCGGACGTA
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7151    GGAGATCTCA TCGCCAGGGT GCCAGCCCCC TGCCCAGCGG CTGTGCCAGC
            CCTCTAGAGT AGCGGTCCCA CGGTCGGGGG ACGGGTCGCC GACACGGTCG
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7201    CTGTGCTACC CAGCCCAGCC TGCCAGCTGG TTCTGCACCA GATACTGAAG
            GACACGATGG GTCGGGTCGG ACGGTCGACC AAGACGTGGT CTATGACTTC
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7251    GGTGGCTCGG GGACATACTG CCTCAATGTG TCTCTGGCTG ATACCAACAG
            CCACCGAGCC CCTGTATGAC GGAGTTACAC AGAGACCGAC TATGGTTGTC
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7301    CCTGGCAGTG GTCAGCACCC AGCTTATCAT GCCTGGTCAA GAAGCAGGCC
            GGACCGTCAC CAGTCGTGGG TCGAATAGTA CGGACCAGTT CTTCGTCCGG
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7351    TTGGGCAGGT TCCGCTGATC GTGGGCATCT TGCTGGTGTT GATGGCTGTG
            AACCCGTCCA AGGCGACTAG CACCCGTAGA ACGACCACAA CTACCGACAC
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7401    GTCCTTGCAT CTCTGATATA TAGGCGCAGA CTTATGAAGC AAGACTTCTC
            CAGGAACGTA GAGACTATAT ATCCGCGTCT GAATACTTCG TTCTGAAGAG
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7451    CGTACCCCAG TTGCCACATA GCAGCAGTCA CTGGCTGCGT CTACCCCGCA
            GCATGGGGTC AACGGTGTAT CGTCGTCAGT GACCGACGCA GATGGGGCGT
                              gp100(M)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7501    TCTTCTGCTC TTGTCCCATT GGTGAGAACA GCCCCCTCCT CAGTGGGCAG
            AGAAGACGAG AACAGGGTAA CCACTCTTGT CGGGGGAGGA GTCACCCGTC
            gp100(M)                          42K promoter
            ~~~~~~~~~                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
    7551    CAGGTCTGAT TTTTATTCTA GTTCAAAAAA ATATAAATGA TTCACCATCT
            GTCCAGACTA AAAATAAGAT CAAGTTTTTT TATATTTACT AAGTGGTAGA
```

```
                          42K promoter
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7601    GATAGAAAAA AAATTTATTG GGAGAATATG ATAATATTTT GGGATTTCAA
                CTATCTTTTT TTTAAATAAC CCTCTTATAC TATTATAAAA CCCTAAAGTT
                       42K promoter                          Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7651    AATTGAAAAT ATATAATTAC AATATAAATC TAGACCACCA TGCCAAGAGA
                TTAACTTTTA TATATTAATG TTATATTTAG ATCTGGTGGT ACGGTTCTCT
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7701    AGATGCTCAC TTCATCTATG GTTACCCCAA GAAGGGGCAC GGCCACTCTT
                TCTACGAGTG AAGTAGATAC CAATGGGGTT CTTCCCCGTG CCGGTGAGAA
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7751    ACACCACGGC TGAAGAGGCC GCTGGGATCG GCATCCTGAC AGTGATCCTG
                TGTGGTGCCG ACTTCTCCGG CGACCCTAGC CGTAGGACTG TCACTAGGAC
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7801    GGAGTCTTAC TGCTCATCGG CTGTTGGTAT TGTAGAAGAC GAAATGGATA
                CCTCAGAATG ACGAGTAGCC GACAACCATA ACATCTTCTG CTTTACCTAT
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7851    CAGAGCCTTG ATGGATAAAA GTCTTCATGT TGGCACTCAA TGTGCCTTAA
                GTCTCGGAAC TACCTATTTT CAGAAGTACA ACCGTGAGTT ACACGGAATT
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7901    CAAGAAGATG CCCACAAGAA GGGTTTGATC ATCGGGACAG CAAAGTGTCT
                GTTCTTCTAC GGGTGTTCTT CCCAAACTAG TAGCCCTGTC GTTTCACAGA
                                   Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        7951    CTTCAAGAGA AAAACTGTGA ACCTGTGGTT CCCAATGCTC CACCTGCTTA
                GAAGTTCTCT TTTTGACACT TGGACACCAA GGGTTACGAG GTGGACGAAT
                                 Mart-1
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        8001    TGAGAAACTC TCTGCAGAAC AGTCACCACC ACCTTATTCA CCTTAATCTA
                ACTCTTTGAG AGACGTCTTG TCAGTGGTGG TGGAATAAGT GGAATTAGAT
                                             sE/L Promoter
                                   ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        8051    GAGTCGACCT GCAGGCATGC AAAAATTGAA ATTTTATTTT TTTTTTTTGG
                CTCAGCTGGA CGTCCGTACG TTTTTAACTT TAAAATAAAA AAAAAAAACC
        sE/L Promoter
        ~~~~~~~~~~~
                                   Mage 1-3 minigene
                        ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        8101    AATATAAATA ATGGAGTCCT TGCAGCTGGT CTTTGGCATT GACGTGAAGG
                TTATATTTAT TACCTCAGGA ACGTCGACCA GAAACCGTAA CTGCACTTCC
                              Mage 1-3 minigene
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        8151    AAGCAGACCC CACCGGCCAC TCCTATGTCC TTGTCACCTG CCTAGGTCTC
                TTCGTCTGGG GTGGCCGGTG AGGATACAGG AACAGTGGAC GGATCCAGAG
                              Mage 1-3 minigene
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
        8201    TCCTATGATG GCAATAAGCG TAAAGAAGTG GACCCCATCG GCCACTTGTA
                AGGATACTAC CGTTATTCGC ATTTCTTCAC CTGGGGTAGC CGGTGAACAT
```

```
     Mage 1-3 minigene                                        C5 Left Arm
          ~~~~                                                ~~~~~~~
     8251 CTAGTTTTTA TCCCGGGTTT TTATGACTAG TTAATCACGG CCGCTTATAA
          GATCAAAAAT AGGGCCCAAA AATACTGATC AATTAGTGCC GGCGAATATT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8301 AGATCTAAAA TGCATAATTT CTAAATAATG AAAAAAAAGT ACATCATGAG
          TCTAGATTTT ACGTATTAAA GATTTATTAC TTTTTTTTCA TGTAGTACTC
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8351 CAACGCGTTA GTATATTTTA CAATGGAGAT TAACGCTCTA TACCGTTCTA
          GTTGCGCAAT CATATAAAAT GTTACCTCTA ATTGCGAGAT ATGGCAAGAT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8401 TGTTTATTGA TTCAGATGAT GTTTTAGAAA AGAAAGTTAT TGAATATGAA
          ACAAATAACT AAGTCTACTA CAAAATCTTT TCTTTCAATA ACTTATACTT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8451 AACTTTAATG AAGATGAAGA TGACGACGAT GATTATTGTT GTAAATCTGT
          TTGAAATTAC TTCTACTTCT ACTGCTGCTA CTAATAACAA CATTTAGACA
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8501 TTTAGATGAA GAAGATGACG CGCTAAAGTA TACTATGGTT ACAAAGTATA
          AAATCTACTT CTTCTACTGC GCGATTTCAT ATGATACCAA TGTTTCATAT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8551 AGTCTATACT ACTAATGGCG ACTTGTGCAA GAAGGTATAG TATAGTGAAA
          TCAGATATGA TGATTACCGC TGAACACGTT CTTCCATATC ATATCACTTT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8601 ATGTTGTTAG ATTATGATTA TGAAAAACCA AATAAATCAG ATCCATATCT
          TACAACAATC TAATACTAAT ACTTTTTGGT TTATTTAGTC TAGGTATAGA
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8651 AAAGGTATCT CCTTTGCACA TAATTTCATC TATTCCTAGT TTAGAATACT
          TTTCCATAGA GGAAACGTGT ATTAAAGTAG ATAAGGATCA AATCTTATGA
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8701 TTTCATTATA TTTGTTTACA GCTGAAGACG AAAAAAATAT ATCGATAATA
          AAAGTAATAT AAACAAATGT CGACTTCTGC TTTTTTTATA TAGCTATTAT
                          C5 Left Arm
          ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     8751 GAAGATTATG TTAACTCTGC TAATAAGATG AAATTGAATG AGTCTGTGAC
          CTTCTAATAC AATTGAGACG ATTATTCTAC TTTAACTTAC TCAGACACTG
          C5 Left Arm
          ~~~~~
     8801 TGCAGCCAAG CTTGGCACTG GCCGTCGTTT TACAACGTCG TGACTGGGAA
          ACGTCGGTTC GAACCGTGAC CGGCAGCAAA ATGTTGCAGC ACTGACCCTT
     8851 AACCCTGGCG TTACCCAACT TAATCGCCTT GCAGCACATC CCCCTTTCGC
          TTGGGACCGC AATGGGTTGA ATTAGCGGAA CGTCGTGTAG GGGGAAAGCG
     8901 CAGCTGGCGT AATAGCGAAG AGGCCCGCAC CGATCGCCCT TCCCAACAGT
          GTCGACCGCA TTATCGCTTC TCCGGGCGTG GCTAGCGGGA AGGGTTGTCA
     8951 TGCGCAGCCT GAATGGCGAA TGGCGCCTGA TGCGGTATTT TCTCCTTACG
          ACGCGTCGGA CTTACCGCTT ACCGCGGACT ACGCCATAAA AGAGGAATGC
     9001 CATCTGTGCG GTATTTCACA CCGCATATGG TGCACTCTCA GTACAATCTG
          GTAGACACGC CATAAAGTGT GGCGTATACC ACGTGAGAGT CATGTTAGAC
```

```
9051   CTCTGATGCC GCATAGTTAA GCCAGCCCCG ACACCCGCCA ACACCCGCTG
       GAGACTACGG CGTATCAATT CGGTCGGGGC TGTGGGCGGT TGTGGGCGAC
9101   ACGCGCCCTG ACGGGCTTGT CTGCTCCCGG CATCCGCTTA CAGACAAGCT
       TGCGCGGGAC TGCCCGAACA GACGAGGGCC GTAGGCGAAT GTCTGTTCGA
9151   GTGACCGTCT CCGGGAGCTG CATGTGTCAG AGGTTTTCAC CGTCATCACC
       CACTGGCAGA GGCCCTCGAC GTACACAGTC TCCAAAAGTG GCAGTAGTGG
9201   GAAACGCGCG AGACGAAAGG GCCTCGTGAT ACGCCTATTT TTATAGGTTA
       CTTTGCGCGC TCTGCTTTCC CGGAGCACTA TGCGGATAAA AATATCCAAT
9251   ATGTCATGAT AATAATGGTT TCTTAGACGT CAGGTGGCAC TTTTCGGGGA
       TACAGTACTA TTATTACCAA AGAATCTGCA GTCCACCGTG AAAAGCCCCT
9301   AATGTGCGCG GAACCCCTAT TTGTTTATTT TTCTAAATAC ATTCAAATAT
       TTACACGCGC CTTGGGGATA AACAAATAAA AAGATTTATG TAAGTTTATA
9351   GTATCCGCTC ATGAGACAAT AACCCTGATA AATGCTTCAA TAATATTGAA
       CATAGGCGAG TACTCTGTTA TTGGGACTAT TTACGAAGTT ATTATAACTT
                                  Amp(R)
                    ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9401   AAAGGAAGAG TATGAGTATT CAACATTTCC GTGTCGCCCT TATTCCCTTT
       TTTCCTTCTC ATACTCATAA GTTGTAAAGG CACAGCGGGA ATAAGGGAAA
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9451   TTTGCGGCAT TTTGCCTTCC TGTTTTTGCT CACCCAGAAA CGCTGGTGAA
       AAACGCCGTA AAACGGAAGG ACAAAAACGA GTGGGTCTTT GCGACCACTT
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9501   AGTAAAAGAT GCTGAAGATC AGTTGGGTGC ACGAGTGGGT TACATCGAAC
       TCATTTTCTA CGACTTCTAG TCAACCCACG TGCTCACCCA ATGTAGCTTG
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9551   TGGATCTCAA CAGCGGTAAG ATCCTTGAGA GTTTTCGCCC CGAAGAACGT
       ACCTAGAGTT GTCGCCATTC TAGGAACTCT CAAAAGCGGG GCTTCTTGCA
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9601   TTTCCAATGA TGAGCACTTT TAAAGTTCTG CTATGTGGCG CGGTATTATC
       AAAGGTTACT ACTCGTGAAA ATTTCAAGAC GATACACCGC GCCATAATAG
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9651   CCGTATTGAC GCCGGGCAAG AGCAACTCGG TCGCCGCATA CACTATTCTC
       GGCATAACTG CGGCCCGTTC TCGTTGAGCC AGCGGCGTAT GTGATAAGAG
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9701   AGAATGACTT GGTTGAGTAC TCACCAGTCA CAGAAAAGCA TCTTACGGAT
       TCTTACTGAA CCAACTCATG AGTGGTCAGT GTCTTTTCGT AGAATGCCTA
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9751   GGCATGACAG TAAGAGAATT ATGCAGTGCT GCCATAACCA TGAGTGATAA
       CCGTACTGTC ATTCTCTTAA TACGTCACGA CGGTATTGGT ACTCACTATT
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9801   CACTGCGGCC AACTTACTTC TGACAACGAT CGGAGGACCG AAGGAGCTAA
       GTGACGCCGG TTGAATGAAG ACTGTTGCTA GCCTCCTGGC TTCCTCGATT
                               Amp(R)
       ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
9851   CCGCTTTTTT GCACAACATG GGGGATCATG TAACTCGCCT TGATCGTTGG
       GGCGAAAAAA CGTGTTGTAC CCCCTAGTAC ATTGAGCGGA ACTAGCAACC
```

```
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      9901      GAACCGGAGC TGAATGAAGC CATACCAAAC GACGAGCGTG ACACCACGAT
                CTTGGCCTCG ACTTACTTCG GTATGGTTTG CTGCTCGCAC TGTGGTGCTA
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
      9951      GCCTGTAGCA ATGGCAACAA CGTTGCGCAA ACTATTAACT GGCGAACTAC
                CGGACATCGT TACCGTTGTT GCAACGCGTT TGATAATTGA CCGCTTGATG
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     10001      TTACTCTAGC TTCCCGGCAA CAATTAATAG ACTGGATGGA GGCGGATAAA
                AATGAGATCG AAGGGCCGTT GTTAATTATC TGACCTACCT CCGCCTATTT
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     10051      GTTGCAGGAC CACTTCTGCG CTCGGCCCTT CCGGCTGGCT GGTTTATTGC
                CAACGTCCTG GTGAAGACGC GAGCCGGGAA GGCCGACCGA CCAAATAACG
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     10101      TGATAAATCT GGAGCCGGTG AGCGTGGGTC TCGCGGTATC ATTGCAGCAC
                ACTATTTAGA CCTCGGCCAC TCGCACCCAG AGCGCCATAG TAACGTCGTG
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     10151      TGGGGCCAGA TGGTAAGCCC TCCCGTATCG TAGTTATCTA CACGACGGGG
                ACCCCGGTCT ACCATTCGGG AGGGCATAGC ATCAATAGAT GTGCTGCCCC
                                Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
     10201      AGTCAGGCAA CTATGGATGA ACGAAATAGA CAGATCGCTG AGATAGGTGC
                TCAGTCCGTT GATACCTACT TGCTTTATCT GTCTAGCGAC TCTATCCACG
                       Amp(R)
                ~~~~~~~~~~~~~~~~~~~~~~~~
     10251      CTCACTGATT AAGCATTGGT AACTGTCAGA CCAAGTTTAC TCATATATAC
                GAGTGACTAA TTCGTAACCA TTGACAGTCT GGTTCAAATG AGTATATATG
     10301      TTTAGATTGA TTTAAAACTT CATTTTTAAT TTAAAAGGAT CTAGGTGAAG
                AAATCTAACT AAATTTTGAA GTAAAAATTA AATTTTCCTA GATCCACTTC
     10351      ATCCTTTTTG ATAATCTCAT GACCAAAATC CCTTAACGTG AGTTTTCGTT
                TAGGAAAAAC TATTAGAGTA CTGGTTTTAG GGAATTGCAC TCAAAAGCAA
     10401      CCACTGAGCG TCAGACCCCG TAGAAAAGAT CAAAGGATCT TCTTGAGATC
                GGTGACTCGC AGTCTGGGGC ATCTTTTCTA GTTTCCTAGA AGAACTCTAG
     10451      CTTTTTTTCT GCGCGTAATC TGCTGCTTGC AAACAAAAAA ACCACCGCTA
                GAAAAAAAGA CGCGCATTAG ACGACGAACG TTTGTTTTTT TGGTGGCGAT
     10501      CCAGCGGTGG TTTGTTTGCC GGATCAAGAG CTACCAACTC TTTTTCCGAA
                GGTCGCCACC AAACAAACGG CCTAGTTCTC GATGGTTGAG AAAAAGGCTT
     10551      GGTAACTGGC TTCAGCAGAG CGCAGATACC AAATACTGTC CTTCTAGTGT
                CCATTGACCG AAGTCGTCTC GCGTCTATGG TTTATGACAG GAAGATCACA
     10601      AGCCGTAGTT AGGCCACCAC TTCAAGAACT CTGTAGCACC GCCTACATAC
                TCGGCATCAA TCCGGTGGTG AAGTTCTTGA GACATCGTGG CGGATGTATG
     10651      CTCGCTCTGC TAATCCTGTT ACCAGTGGCT GCTGCCAGTG GCGATAAGTC
                GAGCGAGACG ATTAGGACAA TGGTCACCGA CGACGGTCAC CGCTATTCAG
     10701      GTGTCTTACC GGGTTGGACT CAAGACGATA GTTACCGGAT AAGGCGCAGC
                CACAGAATGG CCCAACCTGA GTTCTGCTAT CAATGGCCTA TTCCGCGTCG
     10751      GGTCGGGCTG AACGGGGGGT TCGTGCACAC AGCCCAGCTT GGAGCGAACG
                CCAGCCCGAC TTGCCCCCCA AGCACGTGTG TCGGGTCGAA CCTCGCTTGC
     10801      ACCTACACCG AACTGAGATA CCTACAGCGT GAGCTATGAG AAAGCGCCAC
                TGGATGTGGC TTGACTCTAT GGATGTCGCA CTCGATACTC TTTCGCGGTG
     10851      GCTTCCCGAA GGGAGAAAGG CGGACAGGTA TCCGGTAAGC GGCAGGGTCG
                CGAAGGGCTT CCCTCTTTCC GCCTGTCCAT AGGCCATTCG CCGTCCCAGC
```

```
10901   GAACAGGAGA GCGCACGAGG GAGCTTCCAG GGGGAAACGC CTGGTATCTT
        CTTGTCCTCT CGCGTGCTCC CTCGAAGGTC CCCCTTTGCG GACCATAGAA
10951   TATAGTCCTG TCGGGTTTCG CCACCTCTGA CTTGAGCGTC GATTTTTGTG
        ATATCAGGAC AGCCCAAAGC GGTGGAGACT GAACTCGCAG CTAAAAACAC
11001   ATGCTCGTCA GGGGGGCGGA GCCTATGGAA AAACGCCAGC AACGCGGCCT
        TACGAGCAGT CCCCCCGCCT CGGATACCTT TTTGCGGTCG TTGCGCCGGA
11051   TTTTACGGTT CCTGGCCTTT TGCTGGCCTT TTGCTCACAT GTTCTTTCCT
        AAAATGCCAA GGACCGGAAA ACGACCGGAA AACGAGTGTA CAAGAAAGGA
11101   GCGTTATCCC CTGATTCTGT GGATAACCGT ATTACCGCCT TTGAGTGAGC
        CGCAATAGGG GACTAAGACA CCTATTGGCA TAATGGCGGA AACTCACTCG
11151   TGATACCGCT CGCCGCAGCC GAACGACCGA GCGCAGCGAG TCAGTGAGCG
        ACTATGGCGA GCGGCGTCGG CTTGCTGGCT CGCGTCGCTC AGTCACTCGC
11201   AGGAAGCGGA AGAGCGCCCA ATACGCAAAC CGCCTCTCCC CGCGCGTTGG
        TCCTTCGCCT TCTCGCGGGT TATGCGTTTG GCGGAGAGGG GCGCGCAACC
11251   CCGATTCATT AATGCAGCTG GCACGACAGG TTTCCCGACT GGAAAGCGGG
        GGCTAAGTAA TTACGTCGAC CGTGCTGTCC AAAGGGCTGA CCTTTCGCCC
11301   CAGTGAGCGC AACGCAATTA ATGTGAGTTA GCTCACTCAT TAGGCACCCC
        GTCACTCGCG TTGCGTTAAT TACACTCAAT CGAGTGAGTA ATCCGTGGGG
11351   AGGCTTTACA CTTTATGCTT CCGGCTCGTA TGTTGTGTGG AATTGTGAGC
        TCCGAAATGT GAAATACGAA GGCCGAGCAT ACAACACACC TTAACACTCG
11401   GGATAACAAT TTCACACAGG AAACAGCTAT GACCATGATT ACGAATTGAA
        CCTATTGTTA AAGTGTGTCC TTTGTCGATA CTGGTACTAA TGCTTAACTT
11451   TTGCGGCCGC AATTCAACGC CGGCGTTAAG
```

FIGURE 6A

NY-ESO-1

Met Gln Ala Glu Gly Arg Gly Thr Gly Gly Ser Thr Gly Asp Ala Asp
Gly Pro Gly Gly Pro Gly Ile Pro Asp Gly Pro Gly Gly Asn Ala Gly
Gly Pro Gly Glu Ala Gly Ala Thr Gly Gly Arg Gly Pro Arg Gly Ala
Gly Ala Ala Arg Ala Ser Gly Pro Gly Gly Gly Ala Pro Arg Gly Pro
His Gly Gly Ala Ala Ser Gly Leu Asn Gly Cys Cys Arg Cys Gly Ala
Arg Gly Pro Glu Ser Arg Leu Leu Glu Phe Tyr Leu Ala Met Pro Phe
Ala Thr Pro Met Glu Ala Glu Leu Ala Arg Arg Ser Leu Ala Gln Asp
Ala Pro Pro Leu Pro Val Pro Gly Val Leu Leu Lys Glu Phe Thr Val
Ser Gly Asn Ile Leu Thr Ile Arg Leu Thr Ala Ala Asp His Arg Gln
Leu Gln Leu Ser Ile Ser Ser Cys Leu Gln Gln Leu Ser Leu Leu Met
Trp Ile Thr Gln Cys Phe Leu Pro Val Phe Leu Ala Gln Pro Pro Ser
Gly Gln Arg Arg

FIGURE 6C

TRP-2

Met Ser Pro Leu Trp Trp Gly Phe Leu Leu Ser Cys Leu Gly Cys Lys Ile
Leu Pro Gly Ala Gln Gly Gln Phe Pro Arg Val Cys Met Thr Val Asp Ser
Leu Val Asn Lys Glu Cys Cys Pro Arg Leu Gly Ala Glu Ser Ala Asn Val
Cys Gly Ser Gln Gln Gly Arg Gly Gln Cys Thr Glu Val Arg Ala Asp Thr
Arg Pro Trp Ser Gly Pro Tyr Ile Leu Arg Asn Gln Asp Asp Arg Glu Leu
Trp Pro Arg Lys Phe Phe His Arg Thr Cys Lys Cys Thr Gly Asn Phe Ala
Gly Tyr Asn Cys Gly Asp Cys Lys Phe Gly Trp Thr Gly Pro Asn Cys Glu
Arg Lys Lys Pro Pro Val Ile Arg Gln Asn Ile His Ser Leu Ser Pro Gln
Glu Arg Glu Gln Phe Leu Gly Ala Leu Asp Leu Ala Lys Lys Arg Val His
Pro Asp Tyr Val Ile Thr Thr Gln His Trp Leu Gly Leu Leu Gly Pro Asn
Gly Thr Gln Pro Gln Phe Ala Asn Cys Ser Val Tyr Asp Phe Phe Val Trp
Leu His Tyr Tyr Ser Val Arg Asp Thr Leu Leu Gly Pro Gly Arg Pro Tyr
Arg Ala Ile Asp Phe Ser His Gln Gly Pro Ala Phe Val Thr Trp His Arg
Tyr His Leu Leu Cys Leu Glu Arg Asp Leu Gln Arg Leu Ile Gly Asn Glu
Ser Phe Ala Leu Pro Tyr Trp Asn Phe Ala Thr Gly Arg Asn Glu Cys Asp
Val Cys Thr Asp Gln Leu Phe Gly Ala Ala Arg Pro Asp Asp Pro Thr Leu
Ile Ser Arg Asn Ser Arg Phe Ser Ser Trp Glu Thr Val Cys Asp Ser Leu
Asp Asp Tyr Asn His Leu Val Thr Leu Cys Asn Gly Thr Tyr Glu Gly Leu
Leu Arg Arg Asn Gln Met Gly Arg Asn Ser Met Lys Leu Pro Thr Leu Lys
Asp Ile Arg Asp Cys Leu Ser Leu Gln Lys Phe Asp Asn Pro Pro Phe Phe
Gln Asn Ser Thr Phe Ser Phe Arg Asn Ala Leu Glu Gly Phe Asp Lys Ala
Asp Gly Thr Leu Asp Ser Gln Val Met Ser Leu His Asn Leu Val His Ser
Phe Leu Asn Gly Thr Asn Ala Leu Pro His Ser Ala Ala Asn Asp Pro Ile
Phe Val Val Leu His Ser Phe Thr Asp Ala Ile Phe Asp Glu Trp Met Lys
Arg Phe Asn Pro Pro Ala Asp Ala Trp Pro Gln Glu Leu Ala Pro Ile Gly
His Asn Arg Met Tyr Asn Met Val Pro Phe Phe Pro Pro Val Thr Asn Glu
Glu Leu Phe Leu Thr Ser Asp Gln Leu Gly Tyr Ser Tyr Ala Ile Asp Leu
Pro Val Ser Val Glu Glu Thr Pro Gly Trp Pro Thr Thr Leu Leu Val Val
Met Gly Thr Leu Val Ala Leu Val Gly Leu Phe Val Leu Leu Ala Phe Leu
Gln Tyr Arg Arg Leu Arg Lys Gly Tyr Thr Pro Leu Met Glu Thr His Leu
Ser Ser Lys Arg Tyr Thr Glu Glu Ala

FIGURE 6D

gp100 and gp100M

```
1         MDL VLKRCLLHLA VIGALLAVGA TKVPRNQDWL GVSRQLRTKA WNRQLYPEWT
2         *** ********** ********** ********** ********** **********

1 EAQRLDCWRG GQVSLKVSND GPTLIGANAS FSIALNFPGS QKVLPDGQVI WVNNTIINGS
2 ********** ********** ********** ********** ********** **********

1 QVWGGQPVYP QETDDACIFP DGGPCPSGSW SQKRSFVYVW KTWGQYWQFL GGPVSGLSIG
2 ********** ********** ********** ********** ********V* **********

1 TGRAMLGTHT MEVTVYHRRG SRSYVPLAHS SSAFTITDQV PFSVSVSQLR ALDGGNKHFL
2 ********** ********** ********** ******M*** ********** **********

1 RNQPLTFALQ LHDPSGYLAE ADLSYTWDFG DSSGTLISRA LVVTHTYLEP GPVTAQVVLQ
2 ********** ********** ********** ********** ********** ****V*****

1 AAIPLTSCGS SPVPGTTDGH RPTAEAPNTT AGQVPTTEVV GTTPGQAPTA EPSGTTSVQV
2 ********** ********** ********** ********** ********** **********

1 PTTEVISTAP VQMPTAESTG MTPEKVPVSE VMGTTLAEMS TPEATGMTPA EVSIVVLSGT
2 ********** ********** ********** ********** ********** **********

1 TAAQVTTTEW VETTARELPI PEPEGPDASS IMSTESITGS LGPLLDGTAT LRLVKRQVPL
2 ********** ********** ********** ********** ********** **********

1 DCVLYRYGSF SVTLDIVQGI ESAEILQAVP SGEGDAFELT VSCQGGLPKE ACMEISSPGC
2 ********** ********** ********** ********** ********** **********

1 QPPAQRLCQP VLPSPACQLV LHQILKGGSG TYCLNVSLAD TNSLAVVSTQ LIMPGQEAGL
2 ********** ********** ********** ********** ********** **********

1 GQVPLIVGIL LVLMAVVLAS LIYRRRLMKQ DFSVPQLPHS SSHWLRLPRI FCSCPIGENS
2 ********** ********** ********** ********** ********** **********

1 PLLSGQQV2 ********

Key
*=identical amino acid residue
1=gp100
2=gp100M
```

FIGURE 6E

MART-1

Met Pro Arg Glu Asp Ala His Phe Ile Tyr Gly Tyr Pro
Lys Lys Gly His Gly His Ser Tyr Thr Thr Ala Glu Glu
Ala Ala Gly Ile Gly Ile Leu Thr Val Ile Leu Gly Val
Leu Leu Leu Ile Gly Cys Trp Tyr Cys Arg Arg Arg Asn
Gly Tyr Arg Ala Leu Met Asp Lys Ser Leu His Val Gly
Thr Gln Cys Ala Leu Thr Arg Arg Cys Pro Gln Glu Gly
Phe Asp His Arg Asp Ser Lys Val Ser Leu Gln Glu Lys
Asn Cys Glu Pro Val Val Pro Asn Ala Pro Pro Ala Tyr
Glu Lys Leu Ser Ala Glu Gln Ser Pro Pro Pro Tyr Ser
Pro

FIGURE 6F

MAGE-1

Met Ser Asp Asn Lys Lys Pro Asp Lys Ala His Ser Gly Ser Gly Gly
Asp Gly Asp Gly Asn Arg Cys Asn Leu Leu His Arg Tyr Ser Leu Glu
Glu Ile Leu Pro Tyr Leu Gly Trp Leu Val Phe Ala Val Val Thr Thr
Ser Phe Leu Ala Leu Gln Met Phe Ile Asp Ala Leu Tyr Glu Glu Gln
Tyr Glu Arg Asp Val Ala Trp Ile Ala Arg Gln Ser Lys Arg Met Ser
Ser Val Asp Glu Asp Glu Asp Asp Glu Asp Asp Glu Asp Asp Tyr Tyr
Asp Asp Glu Asp Asp Asp Asp Asp Ala Phe Tyr Asp Asp Glu Asp Asp
Glu Glu Glu Glu Leu Glu Asn Leu Met Asp Asp Glu Ser Glu Asp Glu
Ala Glu Glu Glu Met Ser Val Glu Met Gly Ala Gly Ala Glu Glu Met
Gly Ala Gly Ala Asn Cys Ala Cys Val Pro Gly His His Leu Arg Lys
Asn Glu Val Lys Cys Arg Met Ile Tyr Phe Phe His Asp Pro Asn Phe
Leu Val Ser Ile Pro Val Asn Pro Lys Glu Gln Met Glu Cys Arg Cys
Glu Asn Ala Asp Glu Glu Val Ala Met Glu Glu Glu Glu Glu Glu Glu
Glu Glu Glu Glu Glu Glu Glu Met Gly Asn Pro Asp Gly Phe Ser Pro

FIGURE 6G

MAGE-3

mpleqrsqhc kpeeglearg ealglvgaqa pateeqeaas ssstlvevtl gevpaaespd
ppqspqgass lpttmnyplw sqsyedssnq eeegpstfpd lesefqaals rkvaelvhfl
llkyrarepv tkaemlgsvv gnwqyffpvi fskassslql vfgielmevd pighlyifat
clglsydgll gdnqimpkag lliivlaiia regdcapeek iweelsvlev fegredsilg
dpkkllтqhf vqenyleyrq vpgsdpacye flwgpralve tsyvkvlhhm vkisggphis
ypplhewvlr egee

FIGURE 6H
B7.1

mghtrrqgts pskcpylnff qllvlaglsh fcsgvihvtk evkevatlsc ghnvsveela
qtriywqkek kmvltmmsgd mniwpeyknr tifditnnls ivilalrpsd egtyecvvlk
yekdafkreh laevtlsvka dfptpsisdf eiptsnirri icstsggfpe phlswlenge
elnainttvs qdpetelyav sskldfnmtt nhsfmcliky ghlrvnqtfn wnttkqehfp
dnllpswait lisvngifvi ccltycfapr crerrrnerl rresvrpv

FIGURE 6I
LFA-3

mvagsdagra lgvlsvvcll hcfgfiscfs qqiygvvygn vtfhvpsnvp lkevlwkkqk
dkvaelense frafssfknr vyldtvsgsl tiynltssde deyemespni tdtmkfflyv
leslpsptlt caltngsiev qcmipehyns hrglimyswd cpmeqckrns tsiyfkmend
lpqkiqctls nplfnttssi ilttcipssg hsrhryalip iplavittci vlymngilkc
drkpdrtnsn

FIGURE 6J
ICAM-1*

mapssprpal pallvllgal fpgpgnaqts vspskvilpr ggsvlvtcst scdqpkllgi
etplpkkell lpgnnrkvye lsnvqedsqp mcysncpdgq staktfltvy wtpervelap
lpswqpvgkn ltlrcqvegg apranltvvl lrgekelkre pavgepaevt ttvlvrrdhh
ganfscrtel dlrpqglelf entsapyqlq tfvlpatppq lvsprvlevd tqgtvvcsld
glfpvseaqv hlalgdqrln ptvtygndsf sakasvsvta edegtqrltc avilgnqsqe
tlqtvtiysf papnviltkp evsegtevtv kceahprakv tlngvpaqpl gpraqlllka
tpedngrsfs csatlevagq lihknqtrel rvlygprlde rdcpgnwtwp ensqqtpmcq
awgnplpelk clkdgtfplp igesvtvtrd legtylcrar stqgevtrev tvnvlsprye
iviitvvaaa vimgtaglst ylynrqrkik kyrlqqaqkg tpmkpntqat pp

*mature sequence begins at residue 28 (q)